

THE PLASMA PROTEINS AND CARDIAC OEDEMA.

THESIS

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by

WILLIAM A. R. THOMSON, M.B., Ch.B.,

Surgeon Lieutenant,

Royal Navy.

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ψυχῆς γὰρ θάνατος ὕδωρ γένεσθαι,  
ὕδαρ δὲ θάνατος γῆν γένεσθαι·  
ἐκ γῆς δὲ ὕδωρ γίνεσθαι,  
ἐξ ὕδατος δὲ ψυχή.

Heraclitus of Ephesus on Nature.

LXVIII.

## INTRODUCTION.

Since ancient times dropsy has been one of the most dreaded of the "ills to which man is heir". To the lay mind it has suggested something inevitable and almost supernatural, while to the physician it has involved the utilisation of the most drastic measures in his therapeutic armamentarium, measures which, drastic though they might be, all too often failed in their purpose. With the happy chance that introduced WITHERING (155) to the "old wife's tale" and so made the foxglove one of the most potent means of combating this enemy of man, marked progress was made in the amelioration of the lot of those who suffered from cardiac oedema. In spite, however, of much intensive work a complete understanding of the factors responsible for the production of oedema, including that of cardiac origin, has yet to be found. One of the most fruitful lines of research of recent years has been the investigation of the rôle of the plasma proteins in the causation of oedema.

Probably one of the earliest recorded cases of oedema due to plasma protein deficiency is that of Heraclitus /



Heraclitus, one of the Ionian school of philosophy, who, as recorded by DIOGENES, died a miserable death, waterlogged and oedematous, as a result apparently of retiring to the mountains and existing merely upon grass and herbs. To give DIOGENES' own graphic description (35):-

Καὶ γέλος μσανθρωπήσας καὶ  
ἐκπαρήσας ἐν νοτς ὄρεσι δεητὰ το, πῶς  
σιτοῦμένος καὶ βονάνας. καὶ μέντοι καὶ  
διὰ τοῦτο περὶ τραπέζης εἰς ὕδρον  
κατηλθὲν εἰς ἄστυ καὶ τῶν λατρῶν  
αἰνυγματοῦς ἐπυνθάνετο ἐλύναι  
ἐξ ἐπομβρίας αὐχμὸν πολήσας τῶν  
δὲ μὴ συνδέντων, αὐτὸν εἰς  
βουτράλου κατορύξας τῇ τῶν βολίων  
ἀλῆα ἤλπισεν ἐξαρμοσθήσεσθαι.  
οὐδὲν δ' ἀνύων οὐδ' οὕτως, ἐτελεύτα  
βλοῦς ἐν γ' ἐξήκουα.

Καὶ ἔσται εἰς αὐτὸν ἡμῶν  
οὕτως ἔχον.

πολλὰ κς ἡράκλειτον ἐθαύμασα, πῶς  
ποτὲ τοῦ ζῆν  
ᾧ δ' εὐακλήσας δνιμορος εἶεν ἔθανεν  
σῶμα γὰρ ἀρδένσασα κακὴ νόσος  
ἔτανε φέγγος  
ἔσβεσεν ἐν βλεφάροις καὶ σκοτον  
ηγάγετο.

It was RICHARD BRIGHT, however, who, with that amazing perspicacity which has given his work a permanent position in the annals of medicine, first drew attention to the fact that the oedema of the disease now named after him was accompanied by a deficiency in the plasma proteins. "If the blood is drawn it is often buffed, or the serum is milky and opaque; and nice analysis will frequently detect a great deficiency of albumin".(20) The significance of this fact was not appreciated for many years, and it was almost a century later that EPSTEIN (42) published the first of a series of papers in which he finally showed that this plasma protein deficiency was one of the principal factors in the causation of renal oedema.

Since EPSTEIN's original publication in 1912 intensive studies of the plasma proteins have been made in many diseases by workers in America, on the Continent and in this country, and included in these studies have been cases of cardiac oedema. Many such cases have been studied on the Continent, and in 1932 PAYNE and PETERS (117) in America published the results of their studies on twenty-four patients with heart disease in various stages of decompensation. In this country no such study has yet been published, although isolated cases /

cases have been referred to in the course of papers on other forms of oedema, and it is the purpose of this study to give the results of an investigation into the plasma proteins in a series of patients suffering from heart disease both with and without oedema. It should be borne in mind that, definite though the results are, it is not suggested for one moment that the plasma protein deficiency which has been found to exist in cardiac oedema is the entire cause of the oedema in such cases, but it is suggested that such a deficiency is one of the important factors in the causation of this very common condition.

#### METHODS /

### METHODS.

Ten c.c. of blood were withdrawn from the antecubital vein, care being taken to avoid stasis or haemolysis. Having been oxalated the specimen was centrifuged within a quarter of an hour, and all estimations were carried out on the plasma. At regular intervals, duplicate estimations were done in order to ensure accuracy, but at no time was it ever found necessary to discard a result because of lack of correlation between duplicate samples. Any sample that had undergone haemolysis was discarded as being invalid for accurate estimation. The actual estimation was carried out immediately after centrifuging in the vast majority of cases. Occasionally, owing to the exigencies of hospital routine, this was not possible, and in such cases the estimation was carried as far as the stage of the microkjeldahl process, and then laid aside for a few hours.

The plasma having been obtained, 1 c.c., 2 c.c. and 4 c.c. were accurately pipetted into volumetric flasks of respectively 10 c.c. 20 c.c. and 20 c.c. capacity. The plasma in the 10 c.c. flask was made up to volume with distilled water, shaken well and then 1 c.c. of the diluted plasma (equivalent to 0.1 c.c. of plasma) was taken and its total nitrogen estimated /

estimated by the ordinary microkjeldahl process.

In this way the total nitrogen content of the specimen was obtained.

To the 20 c.c. flask containing 2 c.c. of plasma were added a few c.c. of distilled water, 2 c.c. of 10 per cent. Sodium Tungstate, and 2 c.c. of  $\frac{2}{3}$  N. Sulphuric Acid, thus precipitating the proteins. Distilled water was then added to the 20 c.c. mark, the whole shaken and filtered. Of the filtrate 10 c.c. were taken (equivalent to 1 c.c. of plasma) and the nitrogen content again estimated by the microkjeldahl method, thus obtaining the non-protein nitrogen content of the specimen.

In the remaining 20 c.c. flask sufficient solid Magnesium Sulphate was placed to saturate the 4 c.c. of plasma, and the sample then made up to volume with a saturated solution of Magnesium Sulphate, thus precipitating the globulin. The specimen was then shaken well, allowed to stand for a time and filtered. 1 c.c. of the filtrate (equivalent to 0.2 c.c. of plasma) was then used to estimate the nitrogen content by means of the microkjeldahl method.

By the following simple calculation it was then possible to estimate the amount of globulin, albumin and total protein in the sample:-

Calculation /

Calculation:-

(Total Nitrogen - Non-Globulin Nitrogen) x 6.25  $\equiv$  Globulin.  
(Non-Globulin Nitrogen - Non-Protein Nitrogen) x 6.25  $\equiv$  Albumin.

It was not considered necessary to estimate the fibrinogen separately, and by this method it is included in the figure for the globulin.

The same method was used for estimating the protein content of oedematous fluid, the only difference being that oxalate and centrifuging were not required.

It was originally intended to estimate the colloid osmotic pressure of the blood as well as the protein content, but, in view of the unanimity with which GOVAERTS' figures (57) have been accepted and confirmed, it was finally decided that this was not necessary, and that as accurate results would be obtained by calculating the colloid osmotic pressure from the figures supplied by GOVAERTS.

MATERIAL /



### MATERIAL.

The cases upon which this study is based were all patients in the wards of the Royal Infirmary Edinburgh, under the charge of Professor W. T. Ritchie. Sixty-five cases in all were studied, including twenty-three cases of heart failure with oedema, sixteen cases of heart disease unaccompanied by oedema, eight cases of Bright's disease, five cases of tuberculosis, two cases of ovarian carcinoma with gross ascites, a miscellaneous group of eight cases and three junior members of the hospital staff who were used as "normals". In many of the cases repeated examinations of the plasma proteins were made, so that in all, the plasma proteins were estimated on ninety-one occasions. In addition the protein content of oedematous fluid was estimated on twenty-nine samples.

### RESULTS.



## RESULTS.

### Normals.

Three junior members of the hospital staff, all males in the third decade and all with good health records, were taken as "normals", and the results of the estimations are shown in Table I. If this table be compared with Table II it will be seen that our results are in close agreement with those of other workers. For this reason it was not considered necessary to do a large number of cases in order to establish a normal. The normal total protein content of the plasma is thus 7.12 gm. per cent., of which 4.64 gm. per cent. is albumin and 2.48 gm. per cent. globulin, resulting, according to GOVAERTS' (57) figures, in an osmotic pressure of 39.8 cm. of water. This again is in close agreement with other workers, e.g. GOVAERTS (56), 35-40 cm. water; MEYER (110), 35.5 cm. water (average); IVERSEN and NAKAZAWA (71), 32.5-40.1 cm. water; FELLOWS (47), 32.1-38.0 cm. water; SCHADE and CLAUSEN (129), 31-37 cm. water; MEYERS (106), 40 cm. water; VERNEY (148), 36.7 cm. water. The fact that different workers, using different methods, should have achieved such consistent figures is strong proof in support of the correctness of /

TABLE I.

N O R M A L S.

No.	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
	<u>Per cent.</u>	<u>Per cent.</u>	<u>Per cent.</u>		<u>cm. H<sub>2</sub>O</u>
1.	7.490	4.925	2.565	1.920	42.14
2.	7.126	4.813	2.313	2.081	40.80
3.	6.738	4.175	2.563	1.629	36.48
<u>AVERAGE</u>	7.118	4.637	2.480		
	7.12	4.64	2.48	1.877	39.81

TABLE II.

NORMAL PLASMA PROTEINS.

AUTHORS.	Total Protein	Albumin	Globulin	Alb.: Glo Ratio.
	<u>Gm. per cent.</u>	<u>Gm. per cent.</u>	<u>Gm. per cent.</u>	
MOORE & VAN SLYKE (1930)	7.1	4.3	2.8	1.53
LINDER, LUNDGAARD & VAN SLYKE (1914)	6.73	4.11	2.61	1.57
SALVESEN (1926)	7.0			1.67
THOMSON (1932)	7.12	4.64	2.48	1.88
<u>AVERAGE</u>	6.99	4.35	2.63	1.66

of the figure given for the colloid osmotic pressure of the blood.

#### Cardiac Failure with Oedema.

Thirty-two estimations were made in twenty-three cases of cardiac failure with oedema and the results are shown in Table III and Figure I. From Table III it will be seen that the total protein varied from 3.14 to 7.20 gm. per cent. with an average of 5.28 gm. per cent., the albumin varied from 1.64 to 3.81 gm. per cent., average, 2.69 gm. per cent., while the globulin maximum was 4.38 gm. per cent., the minimum, 1.5 gm. per cent., and the average reading 2.59 gm. per cent. The calculated colloid osmotic pressure varied from 15.32 to 33.97 cm. of water, with an average of 25.32.

Cases O. 230 and O. 163 are of especial interest as in both, the total protein content of the plasma was above 7.0 gm. per cent., while the osmotic pressure was low in both cases. The explanation lies in the fact that in both cases the albumin content was low, while the globulin was well above the normal. This is a typical example of how misleading a simple estimation of the total protein of the blood may be in drawing deductions as to the colloid osmotic pressure.

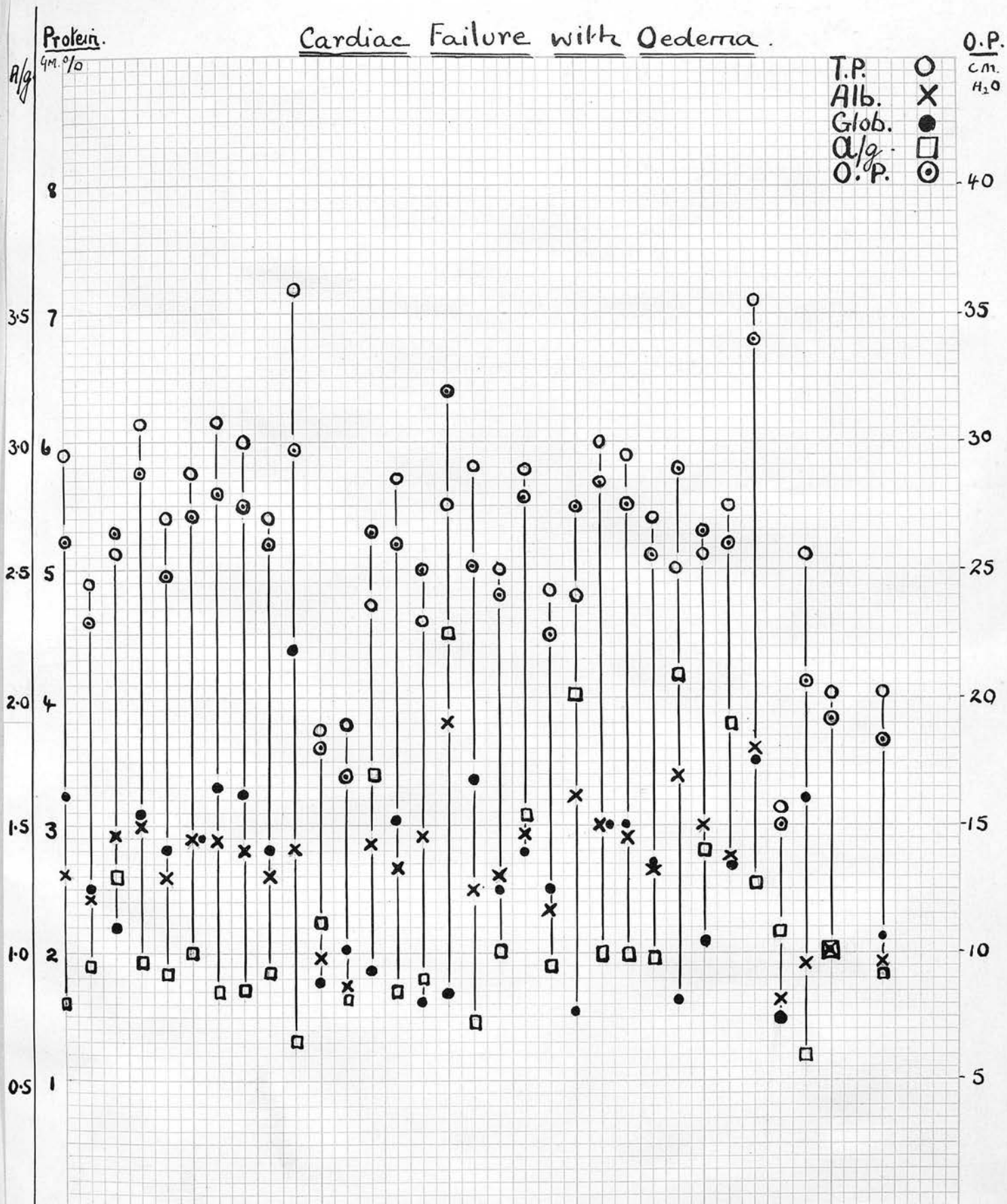
TABLE III.

## CARDIAC FAILURE WITH OEDEMA.

No.	Disease.	Oedema	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
			Per cent.	Per cent.	Per cent.		cm. H <sub>2</sub> O
O. 125	Mitral Stenosis	++++	5.888	2.638	3.250	0.812	26.23
O. 96	Mitral Stenosis Auricular Fibrillation)	+++	4.913	2.413	2.500	0.965	23.07
P. 11	Myocardial Failure	+++ ++	5.163 6.150	2.944 3.025	2.219 3.125	1.327 0.968	26.52 28.90
O. 230	Myocardial Failure	+++ ++ + ++ ++ +++	5.401 5.763 6.163 6.050 5.421 7.200	2.588 2.888 2.850 2.800 2.608 2.825	2.813 2.875 3.313 3.250 2.813 4.375	0.920 1.005 0.860 0.862 0.927 0.646	24.84 27.38 27.95 27.45 25.15 29.74
O. 98	Myocardial Failure	+++ ++ +	3.725 3.775 4.725	1.975 1.713 2.850	1.750 2.062 1.875	1.129 0.831 1.520	18.30 16.94 25.15
O. 149	Myocardial Failure	++++ ++++ ++	5.732 4.550 5.501	2.669 2.925 3.813	3.063 1.625 1.688	0.871 1.800 2.259	26.10 25.22 32.04
G. 83	Mitral Stenosis	++++ ++++ +++	5.863 5.075 5.782	2.488 2.575 2.969	3.375 2.500 2.813	0.737 1.030 1.055	25.34 24.29 27.87
O. 95	Aortic Incompetence) W.R. negative )	Trace +	4.863 4.813	2.363 3.250	2.500 1.563	0.945 2.080	22.69 27.55
N. 11	Mitral Stenosis	+	6.000	3.000	3.000	1.000	28.47
O. 208	Chronic Bronchitis.) Cardiac Failure )	++	5.900	2.900	3.000	0.967	27.72
O. 40	Mitral Stenosis.) ? Subacute Bacterial ) Endocarditis )	++	5.420	2.670	2.750	0.971	25.49
O. 162	Arteriosclerosis ) Cardiac Failure )	+++	5.038	3.413	1.625	2.100	28.90
O. 39	Mitral Stenosis.) Auricular Fibrillation)	+	5.100	3.000	2.100	1.429	26.72
O. 237	Myocardial Failure	Trace	5.525	2.775	2.750	1.009	26.19
O. 163	Myocardial Failure ) Auricular Fibrillation)	++	7.100	3.600	3.500	1.029	33.97
O. 246	Mitral Stenosis	+	3.144	1.644	1.500	1.096	15.32
L. 212	Aortic Incompetence	+	5.138	1.919	3.219	0.596	20.75
O. 155	Carcinoma of Stomach) Arteriosclerosis ) Myocardial Failure )	+++ +++	4.069 4.075	2.038 1.919	2.031 2.516	1.003 0.890	19.33 18.67
AVERAGE			5.282	2.689	2.593	1.114	25.32
MAXIMUM			7.200	3.813	4.375	2.259	33.97
MINIMUM			3.144	1.644	1.500	0.596	15.32



Fig. 1.



A "normal" figure for the total protein may be accompanied by a much diminished osmotic pressure, due to a predominance of the globulin fraction.

MOORE and VAN SLYKE (111) have shown that in glomerulonephritis the "critical" level for the plasma albumin is 2.5 gm. per cent. - if the plasma albumin falls below this level oedema occurs. A study of Table III shows that the "critical" level in cardiac oedema may be taken as 3.2 gm. per cent., for of the cases with cardiac oedema 87.5 per cent. had a plasma albumin content of less than 3.2 gm. per cent. Similarly the "critical" level for the colloid osmotic pressure of the blood is 29 cm. water, 91 per cent. of the cases in this group having a figure below this level.

A further point that may be noted is that there is a considerable degree of correlation between both the plasma albumin and the colloid osmotic pressure on the one hand, and the degree of oedema on the other. This is well illustrated in the three cases, 0.98, 0.149 and 0.230, Figures II, III and IV. A summary of the clinical history of these patients will be found in the Appendix, and it can be noted that in each case, as the protein content of the blood rose, the oedema diminished, the fall in weight being perhaps /



Fig. II.

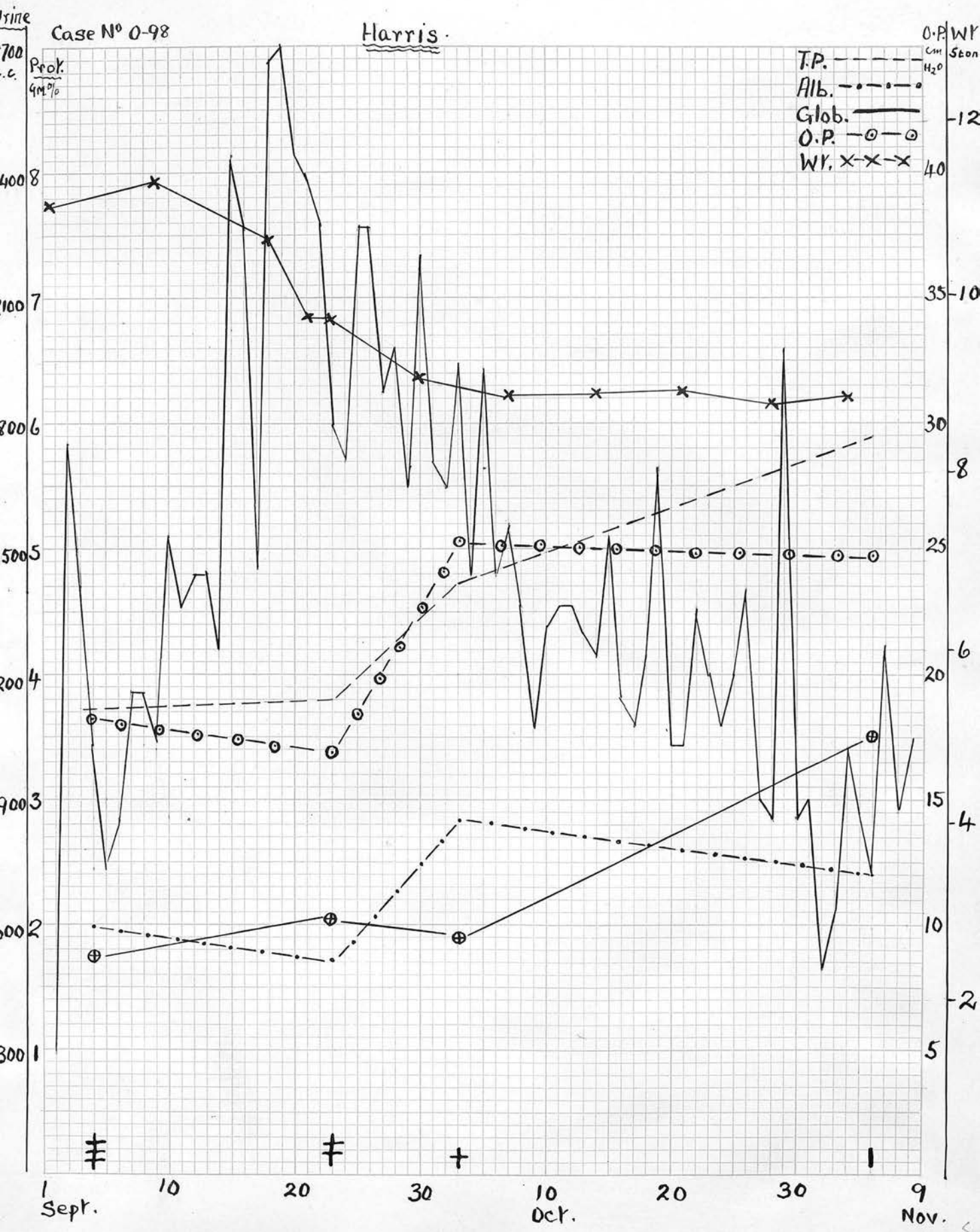
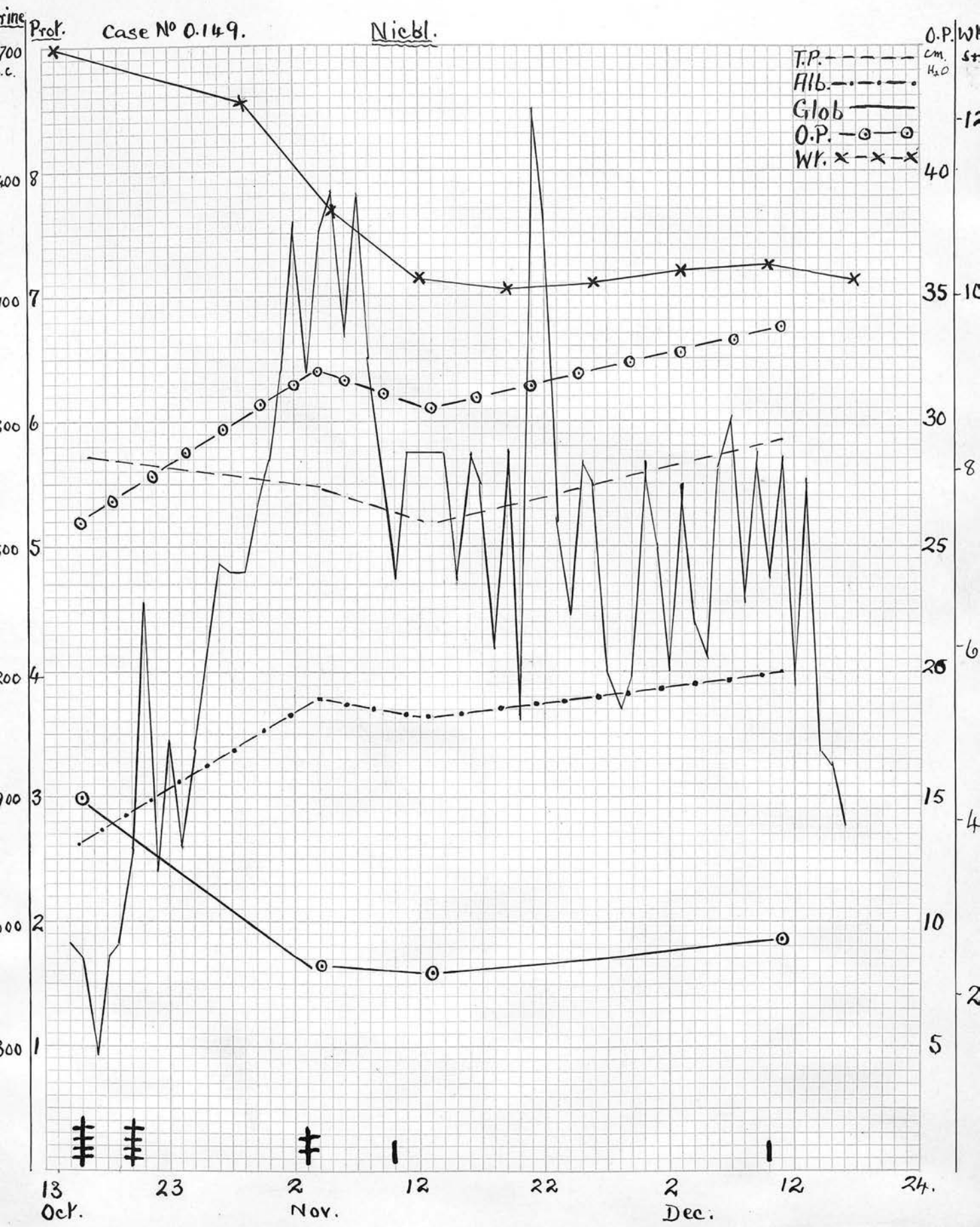


Fig. III.



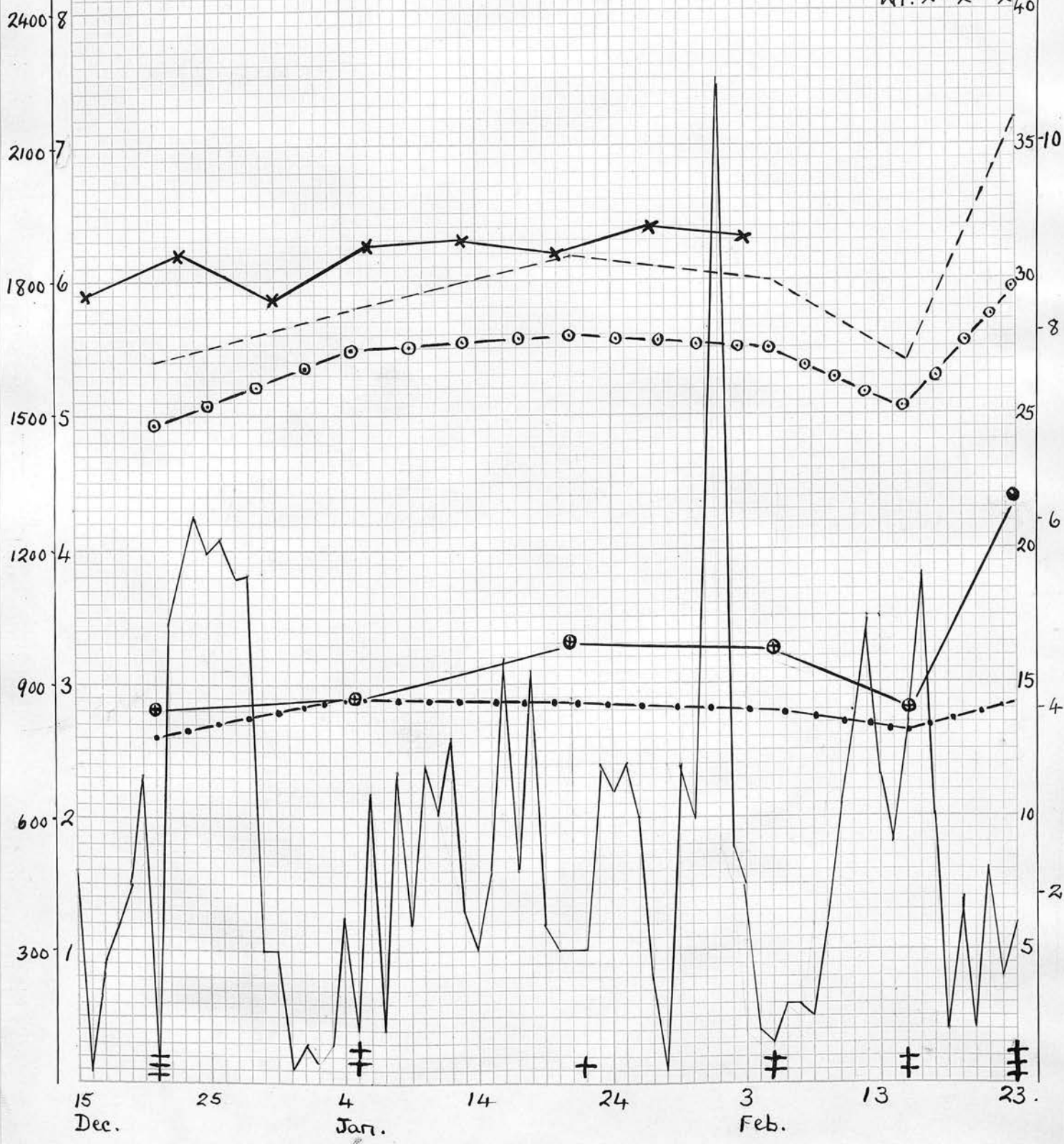
Urine Prof.  
2700 9m. 0.  
c.c.

Case No 0.230.

Dickson.

O.P. WT.  
can. 5T.  
H<sub>2</sub>O

T.P. ---  
Hb. ---  
Glob. ---  
O.P. -o-o-  
Wt. x-x-x-



perhaps the best guide to the disappearance of the oedema. In case 0.98 the colloid osmotic pressure rose from 18.30 to 25.2 cm. water, while the plasma albumin rose from 1.98 to 2.85 gm. per cent., during which time the oedema completely disappeared. A similar course of events occurred with case 0.149, though on a higher level, the colloid osmotic pressure rising from 26.1 cm. of water to 33.9 cm., the corresponding figures for the plasma albumin being 2.67 and 4.01 gm. per cent.; in the meanwhile the oedema had disappeared. Case 0.230 is of particular interest, as here the oedema never disappeared and there were only minor fluctuations in the degree of it. Even so, however, it is evident that with amelioration of the oedema there is a tendency for the colloid osmotic pressure to rise, while a fall in the latter is accompanied by an increase in the degree of oedema, until the final reading when, in spite of a distinct rise in the osmotic pressure, the oedema continued to increase. At the period when this final reading was taken the patient's condition was rapidly becoming worse and nearing a fatal termination. This case would seem to be comparable to those investigated by PETERS and his co-workers (119A) who found, in a group of cases of chronic nephritis who developed /



developed acute heart failure as a terminal condition, that the serum proteins rose sharply while the blood cell volume and oxygen capacity remained unaltered or diminished. They suggest as an explanation that haemoconcentration occurred due to the passage of fluid without protein from the blood to the tissue spaces.

#### Cardiac Disease Without Oedema.

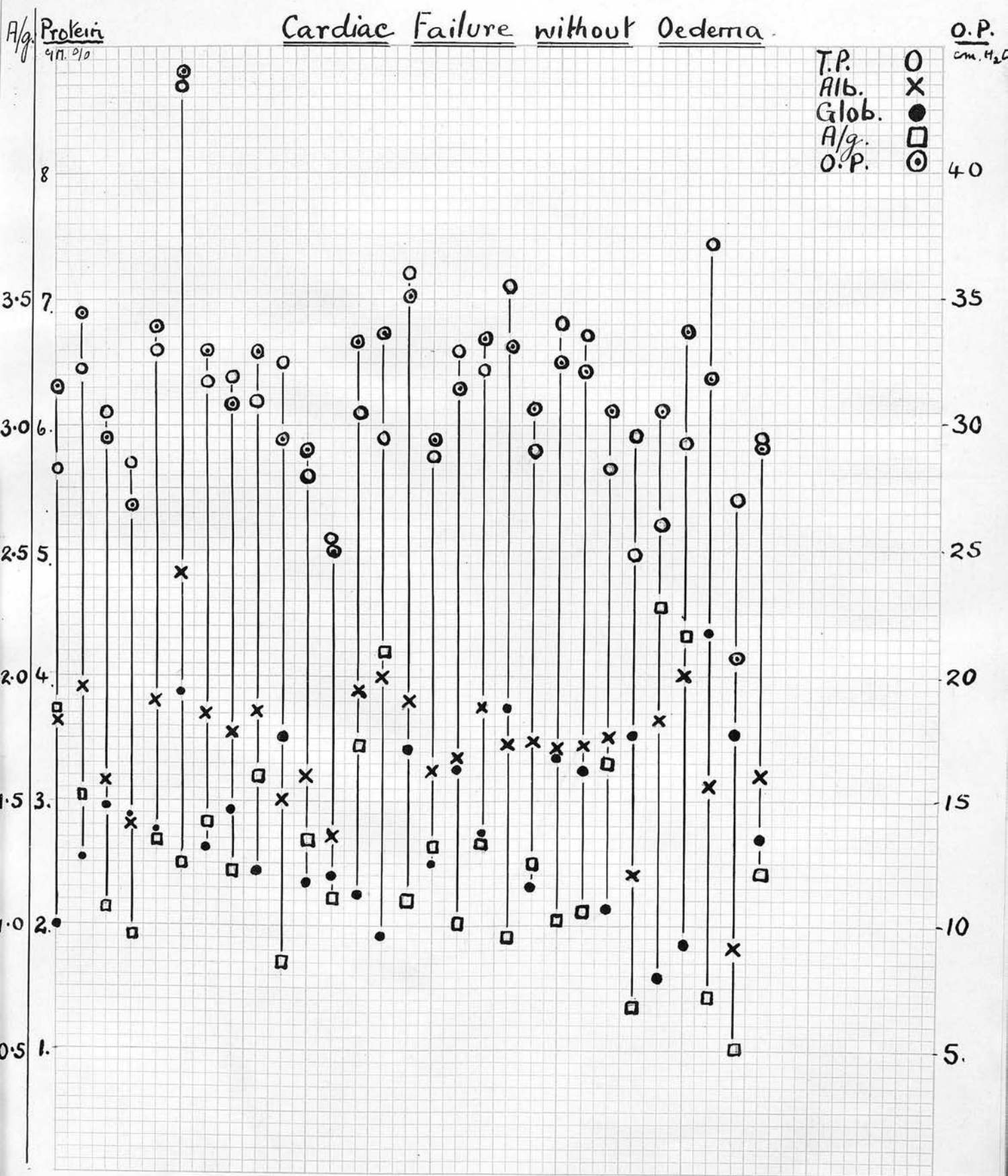
Sixteen such cases were investigated, on whom twenty-nine estimations of the plasma proteins were made. All were admitted to hospital on account of signs and symptoms referable to cardiac involvement. The results are those in Table IV and Figure V from which it can be seen that the average reading for the total plasma protein was 6.34 gm. per cent., the minimum being 5.15 and the maximum 8.71 gm. per cent. The corresponding values for the plasma albumin were 3.56, 2.73 and 4.84 gm. per cent. respectively, while in the case of the colloid osmotic pressure the results were 32.17, 44.03 and 25.31 cm. of water respectively. In several cases the same patient appears in both Table III and Table IV, the reason being that during their stay in hospital the oedema completely disappeared and so they fell into both /

TABLE IV.

CARDIAC FAILURE WITHOUT OEDEMA.

No.	Disease.	Oedema	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
			Per cent.	Per cent.	Per cent.		cm. H <sub>2</sub> O.
O.254	Aortic Incompetence	-	5.663	3.663	2.000	1.832	31.52
G. 59	Myxoedema ) Angina Pectoris )	-	6.476	3.913	2.563	1.527	34.50
O.107	Auricular Fibrillation ) and Flutter )	-	6.126	3.188	2.938	1.085	29.77
O.178	Angina Pectoris	-	5.700	2.825	2.875	0.983	26.91
O.288	Arteriosclerosis	-	6.619	3.838	2.781	1.380	34.36
O.172	Arteriosclerosis	-	8.713	4.838	3.875	1.249	44.03
O.263	Arteriosclerosis	-	6.363	3.738	2.625	1.424	33.30
		-	6.400	3.525	2.875	1.226	31.18
		-	6.213	3.744	2.469	1.516	33.04
K. 89	Osteoarthritis ) Arteriosclerosis )	-	6.519	3.019	3.500	0.863	29.59
G. 29	Mitral Stenosis ) Auricular Fibrillation )	-	5.613	3.238	2.375	1.363	29.05
O.118	Subacute Bacterial ) Endocarditis. ) Aortic Incompetence )	-	5.150	2.731	2.419	1.129	25.31
O.262	Auricular Fibrillation	-	6.138	3.888	2.250	1.728	33.70
O. 42	Chronic Interstitial ) Nephritis. ) Cardiac Failure )	-	5.900	4.000	1.900	2.105	33.87
O. 47	Chronic Interstitial ) Nephritis. ) Cardiac Failure )	-	7.200	3.800	3.400	1.118	35.28
		-	5.781	3.281	2.500	1.312	29.61
O.190	Cerebral Haemorrhage ) Atheroma )	-	6.631	3.350	3.281	1.021	31.66
		-	6.469	3.750	2.719	1.379	33.58
O.204	Arteriosclerosis	-	7.188	3.488	3.750	0.917	33.24
L.251	Arteriosclerosis	-	5.798	3.485	2.313	1.506	30.79
		-	6.800	3.425	3.375	1.015	32.41
		-	6.706	3.456	3.250	1.063	32.40
		-	5.650	3.525	2.125	1.659	30.72
O. 98	Myocardial Failure	-	5.900	2.400	3.500	0.686	24.92
O.149	Myocardial Failure	-	5.238	3.644	1.594	2.286	30.58
		-	5.857	4.013	1.844	2.176	33.85
O.246	Mitral Stenosis	-	7.481	3.106	4.375	0.710	31.95
L.212	Aortic Incompetence	-	5.394	1.831	3.563	0.514	20.75
		-	5.932	3.244	2.688	1.207	29.70
	<u>AVERAGE</u>		6.340	3.555	2.785	1.320	32.17
	<u>MAXIMUM</u>		8.713	4.838	3.875	2.105	44.03
	<u>MINIMUM</u>		5.150	2.731	1.900	0.863	25.31

Fig. V.





both categories - cardiac failure with and without oedema.

Taking the "critical" level for albumin as being 3.2 gm. per cent., it is found that in this group 87 per cent. of the readings are above this value, while with a "critical" level for the colloid osmotic pressure of 29 cm. of water, 91 per cent. of the readings are above the level. It is not suggested that there is anything specific about these "critical" values, but it is nevertheless a striking fact that in the case of the albumin level, 87 per cent. of the cases with oedema are below this level, while 87 per cent. of the cases without oedema are above it, and similarly with the colloid osmotic pressure that 91 per cent. of the cases with oedema are below this level and 91 per cent. of the cases without oedema are above it.

#### Miscellaneous Group.

Eight cases are included in this group, but in only seven were the plasma proteins investigated. The results, recorded in Table V, show that the average value for the total plasma proteins was 6.58 gm. per cent., with a maximum of 7.51 and a minimum of 4.99 gm. per cent. The corresponding values for the plasma /

TABLE V.

M I S C E L L A N E O U S .

No.	Disease	Oedema	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
			<u>Per cent.</u>	<u>Per cent.</u>	<u>Per cent.</u>		<u>cm. H<sub>2</sub>O</u>
M.2	Chronic Myelogenous Leukaemia )	-	7.513	4.263	3.250	1.312	38.48
M.85	Syphilitic Anaemia )	-	7.176	3.238	3.938	0.822	32.09
O.256	Pernicious Anaemia )	-	4.987	3.706	1.281	2.890	30.44
N.259	Chronic Arthritis )	-	6.280	3.590	2.690	1.335	32.31
O.3	Duodenal Ulcer )	-	6.760	4.450	2.310	1.927	38.06
O.244	Diabetes Mellitus )	-	6.244	4.119	2.125	1.938	31.06
O.287	Chronic Bronchitis )	-	7.088	4.463	2.625	1.700	38.77
N.199	Carcinoma of Lung )	Hydro- thorax					
	<u>AVERAGE</u>		6.578	3.975	2.603	1.703	34.46
	<u>MAXIMUM</u>		7.513	4.463	3.938	2.890	38.77
	<u>MINIMUM</u>		4.987	3.238	1.281	0.822	30.44

plasma albumin are 3.98, 4.46, and 3.24 gm. per cent., while for the colloid osmotic pressure they are 34.46, 38.77 and 30.44 cm. of water respectively. Here again the values for both the plasma albumin and the colloid osmotic pressure are all above the "critical" level.

Two of the cases in this group are of interest from the point of view of the relation of the plasma proteins to oedema. The lowest figures both for the plasma proteins and for the osmotic pressure are found in case 0.256 - a case of pernicious anaemia. Oedema is a not uncommon finding in this condition. BRAMWELL (19) in a series of forty-five cases of pernicious anaemia found twenty-three with oedema, and as MAVER (104) has pointed out it is not impossible that a deficiency in the plasma proteins is one of the factors in the causation of this oedema. KYLIN (81) actually found that in these cases the Kolloidosmotie pressure often falls to subnormal levels - even below 200 cm. of water.

EPSTEIN (42) includes diabetes mellitus among the list of diseases in which the globulin is increased while PETERS and his co-workers (120) found reduced plasma proteins regularly in severe diabetes mellitus with chronic malnutrition, and attributed both this reduction /

reduction and the oedema which may occur in diabetes to undernutrition. LABBE and BOULIN (82), in a study of the plasma proteins in diabetes mellitus, reported a moderately high protein content in more than half the cases, and after treatment a fall below normal in many cases. It is thus of interest to note that in case O.244, a comparatively mild case of diabetes mellitus without oedema, the plasma proteins were slightly subnormal.

Attention may also be drawn to the fact that the highest globulin content occurred in a case of syphilitic anaemia (case M.85). This is in accordance with the well known fact that there is a high globulin content in cases of pyogenic and chronic infection. (ERHEN (44A))

#### Tuberculosis.

This is a small group, there being only seven observations on five cases, but it has been kept separate from the previous group as the results are of particular interest from the point of view of this study. The results (Table VI and Figure VI) show that for the group as a whole the average reading for the total plasma protein is 5.40 gm. per cent., with a maximum of 6.26 and a minimum of 4.51 respectively, while /

TABLE VI.

TUBERCULOSIS.

No.	Disease.	Oedema	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
			Per cent.	Per cent.	Per cent.		cm. H <sub>2</sub> O
0.153	Generalised Tuberculosis	Ascites +++	4.506	2.881	1.625	1.773	24.89
		Ascites ++++	4.588	2.588	2.000	1.294	23.41
		Ascites) ++++	5.331	2.706	2.625	1.031	25.52
		Oedema of legs)					
		Ascites) +++	4.632	2.694	1.938	1.390	24.09
		Oedema of legs)					
0.209	Mitral Stenosis } Pulmonary Tuberculosis }	-	6.263	3.200	3.063	1.045	30.10
0.217	Tubercular Pleurisy }	Pleural Effusion) ++	6.263	3.138	3.125	1.004	29.75
N.242	Tubercular Pleurisy	-	6.240	4.180	2.060	2.029	35.53
	<u>AVERAGE</u>		5.403	3.055	2.348	1.367	27.61
	<u>MAXIMUM</u>		6.263	4.180	3.125	2.029	35.53
	<u>MINIMUM</u>		4.506	2.588	1.625	1.004	23.41
<u>Tuberculosis with Gross Ascites.</u> (4 observations)							
	AVERAGE		4.765	2.717	2.047	1.372	24.48
	MAXIMUM		5.331	2.881	2.625	1.773	25.52
	MINIMUM		4.506	2.588	1.625	1.031	23.41
<u>Active Tuberculosis</u> (2 cases)							
	AVERAGE		6.263	3.169	3.094	1.024	29.92
<u>Convalescent Tuberculosis</u> (1 case)							
			6.240	4.180	2.060	2.029	35.53



Fig. VI.

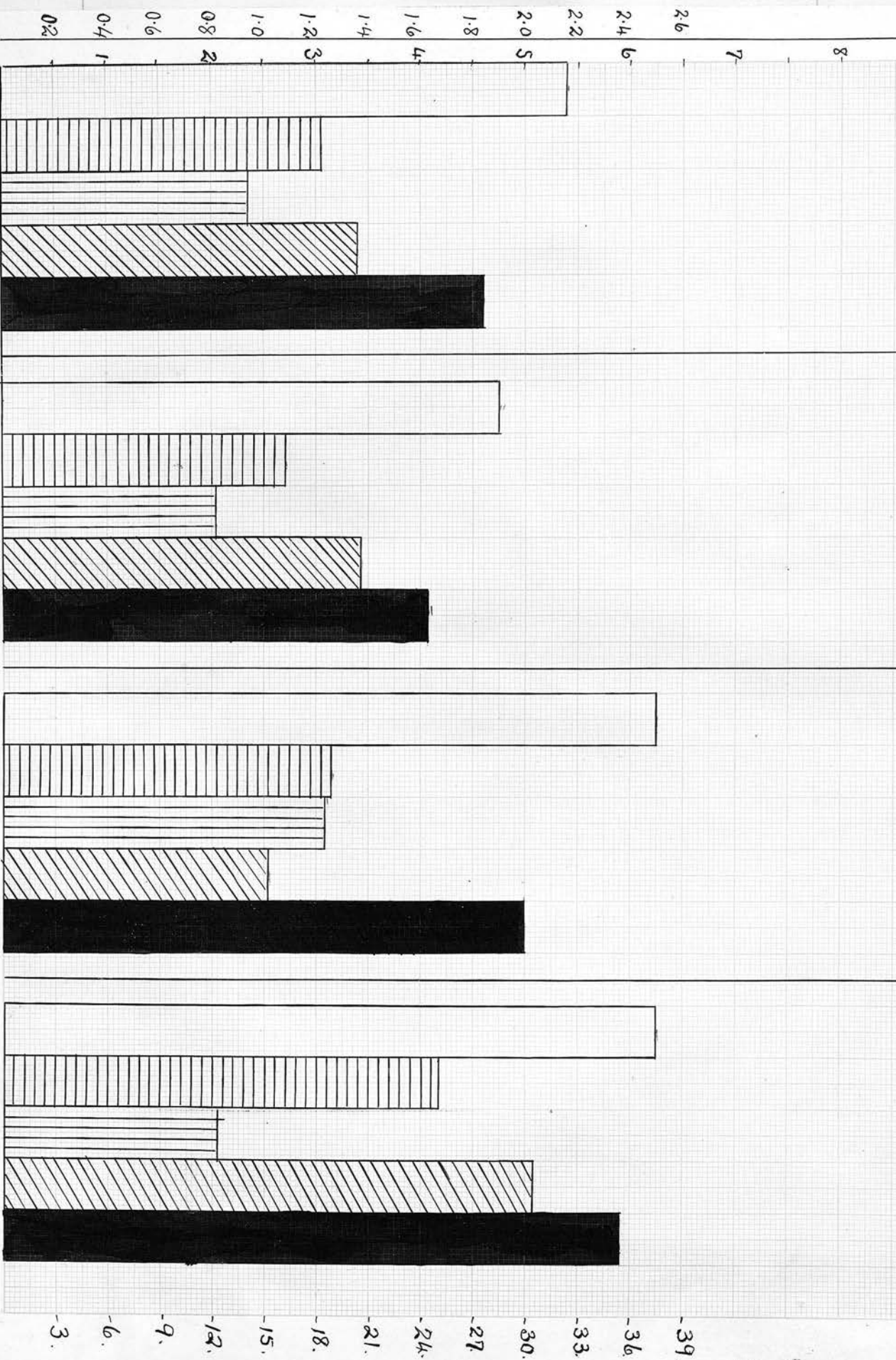
Tuberculosis.

Tuberculosis & Gross Ascites.

Active Tuberculosis.

Convalescent Tuberculosis.

O.P.  
cm H<sub>2</sub>O



while the corresponding values for the plasma albumin are 3.01, 4.18 and 2.59 gm. per cent. respectively, and for the globulin 2.35, 3.13 and 1.63 gm. per cent. respectively.

The division of these cases into four groups as shown in Figure VI reveals several points of interest.

- (i) All cases. There is a fall in the plasma proteins as compared with the normal, the fall being most marked in the albumin fraction with scarcely any drop in the globulin fraction.
- (ii) Tuberculosis with Gross Ascites. There is a still greater fall in the total plasma proteins with a corresponding drop in the colloid osmotic pressure, the protein diminution being almost equally divided between the globulin and albumin fractions, but the albumin-globulin ratio is still 1.37 as compared with 1.88 in the normal group.
- (iii) "Active" Tuberculosis. The total plasma protein figure, though still below that of the normals, attains the highest level of all the cases of Tuberculosis, but when the separate fractions are compared the drop is entirely in the albumin fraction, there being an actual rise in the globulin fraction compared with the normal, resulting /



resulting in an albumin-globulin ratio of 1.02 as against a corresponding figure of 1.88 for the normal group.

- (iv) "Convalescent" Tuberculosis. The total plasma protein is slightly lower than in the "active" group, but the colloid osmotic pressure is the highest of all the subdivisions, 35.53 cm. of water, though still below the normal value. This comparatively high osmotic pressure is due to the preponderance of the albumin fraction, giving an albumin-globulin ratio of 2.03.

To sum up. In all forms of tuberculosis, prior to the convalescent stage, there is a relative or absolute increase in the plasma globulin, this being one of the natural responses of the body to bacterial infection, as has been shown among others by LUTHY (101) and RATHERY and LEVINA (123). Where, however, the disease is accompanied by effusion, e.g. ascites, there is a marked fall in both protein fractions, though that in the globulin fraction is still relatively less, resulting in a corresponding fall in the colloid osmotic pressure. In the convalescent group, where the infection has been arrested or eradicated, there are no effusions and the patient's general nutrition has been improved, the figures return /

return towards normal both in the total protein content of the plasma and in the relative proportions of the two fractions.

#### Renal Oedema.

There were eight cases of glomerulonephritis in this group, on six of which nine observations were made. In the case of the remaining two, observations were only made on the oedema fluid. The results are shown in Table VII and Figure VII. No attempt has been made to group these cases together, but they have been divided into four divisions. In both acute and subacute nephritis unaccompanied by oedema there is only a slight diminution in the plasma protein, particularly in the subacute form, and in the acute form the fall is entirely in the albumin fraction, there being an actual increase in the globulin fraction.

In the cases accompanied by oedema, however, both acute and subacute, there is a distinct fall in the plasma protein, this being much more marked in the subacute form, and once again the fall is most marked in the albumin fraction.

These results are in close agreement with those of other workers and for this reason it was not considered /

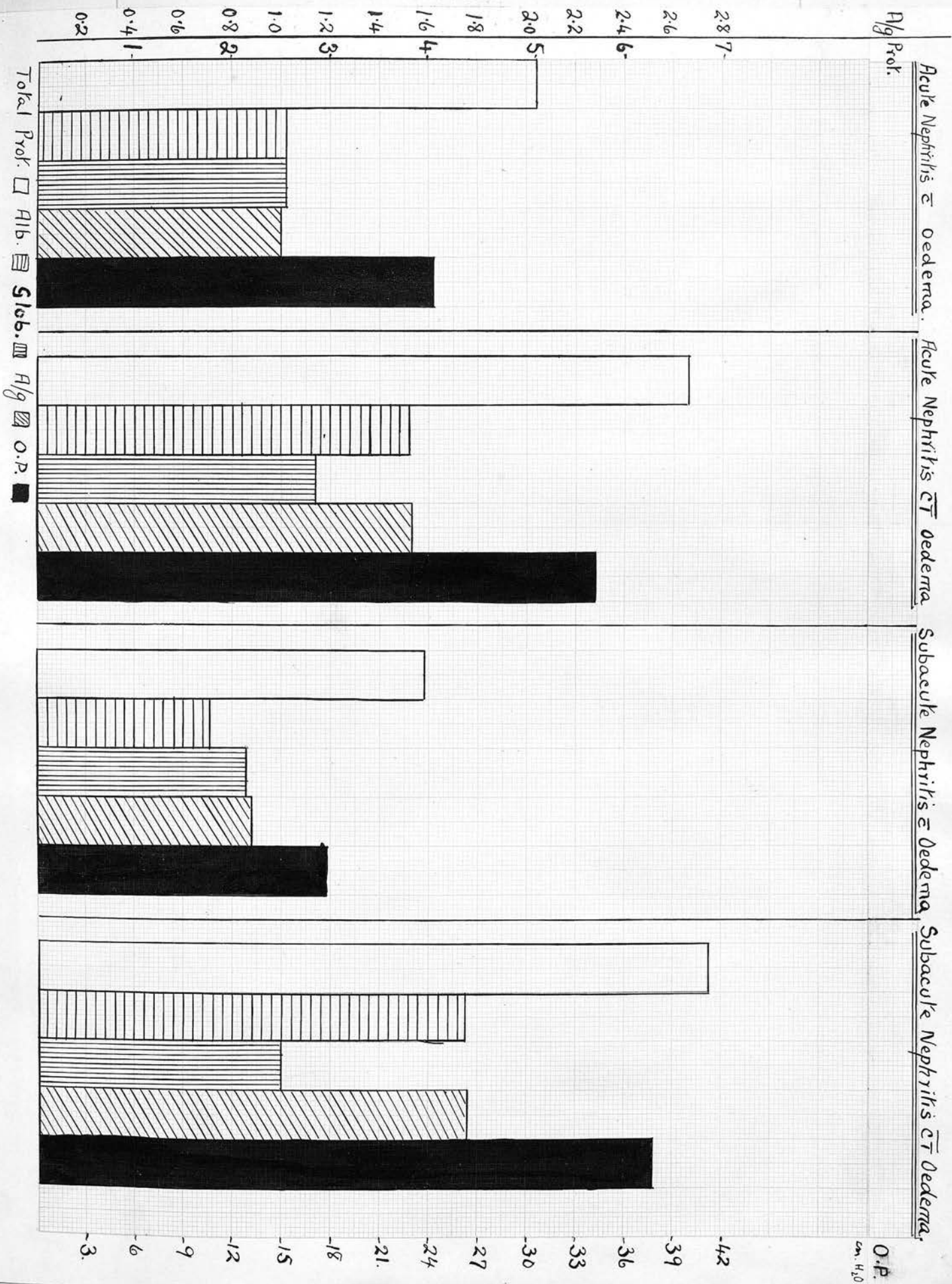
considered necessary to investigate a larger number of cases, as this group was merely acting as a control to the cases of cardiac oedema. EPSTEIN (42) in 1912 reported two cases of "chronic parenchymatous nephritis", both with low plasma protein, the fall in each being entirely in the albumin fraction, the figures being 0.8 and 0.13 gm. per cent. respectively; while in a later publication (44) he gives the average composition of the blood in "chronic parenchymatous nephritis" as total protein, 3.928, albumin, 0.466, and globulin, 3.462 gm. per cent. MENSI (109) states that in children the plasma proteins are below normal in nephritis. ATCHLEY, LOEB, BENEDICT and PALMER (5) found the total proteins to vary from 6.4 to 7.5 gm. per cent. in five cases of acute nephritis, the case with 7.5 gm. per cent. having no oedema, and the case with 6.4 gm. per cent. having most. The same workers investigated fifteen cases of chronic nephritis, and found the total proteins to vary from 4.1 to 7.9 gm. per cent., the lowest figures being in the cases with oedema. GOVAERTS (56) in four cases of "nephritis hydropigenes avec anasarque," found total protein figures of 5.87 - 5.21 gm. per cent. with colloid osmotic pressures of 22 - 12.5 cm. of water. In a later paper (58) he reports on eight cases of glomerulonephritis with oedema with total proteins varying /

TABLE VII.  
R E N A L   O E D E M A.

No.	Disease.	Oedema	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
			Per cent.	Per cent.	Per cent.		cm. H <sub>2</sub> O
O.2	Acute Nephritis	- Haematuria - )	5.900	4.200	1.700	2.471	34.98
O.185	Acute Nephritis	Convalescent	7.751	4.063	3.688	1.102	37.83
H.93	Acute Nephritis	+ Haematuria + )	5.126	2.563	2.563	1.000	24.32
		- Haematuria trace )	6.301	3.113	3.188	0.976	29.69
N.266	Subacute Nephritis )	+++	4.060	2.250	1.810	1.243	20.49
		++	3.800	1.800	2.000	0.900	17.47
		+	3.775	1.775	2.000	0.888	17.28
P.5	Subacute Nephritis ) Uraemia )	++++	4.182	1.369	2.813	0.487	15.81
O.207	Subacute Nephritis )	-	6.875	4.375	2.500	1.750	37.86
	<u>Acute Nephritis with Oedema.</u>		5.126	2.563	2.563	1.000	24.32
	<u>Acute Nephritis without Oedema.</u>						
	AVERAGE		6.651	3.792	2.859	1.516	34.17
	MAXIMUM		7.751	4.200	3.688	2.471	37.83
	MINIMUM		5.900	3.113	1.700	0.976	29.69
	<u>Subacute Nephritis with Oedema.</u>						
	AVERAGE		3.954	1.798	2.156	0.879	17.76
	MAXIMUM		4.182	2.250	2.813	1.243	20.49
	MINIMUM		3.775	1.369	1.810	0.487	15.81
	<u>Subacute Nephritis without Oedema.</u>		6.875	4.375	2.500	1.750	37.86



Fig. VII.





varying from 4.48 to 5.24 gm. per cent. and colloid osmotic pressures of 20 - 25 cm. of water; and six cases of glomerulonephritis without oedema with corresponding figures of 5.15 - 6.92 gm. per cent. and 29.5 - 35 cm. of water.

LINDER, LUNDGAARD, and VAN SLYKE (94), using Volhard and Fahr's classification, found in the nephrotic type of glomerulonephritis that the total proteins were less than 5.5 gm. per cent. in all chronic and subacute cases in which the disease was or had been recently active, and that whenever the total proteins fell below 4 gm. per cent. oedema occurred. On the other hand in the vascular type of glomerulonephritis the total proteins were normal. MOORE and VAN SLYKE (111) in a series of seventy-five cases of nephritis found that no patient with a total protein figure above 5.5 gm. per cent. or albumin above 2.5 gm. per cent. showed any oedema, while all cases with total proteins and albumin below these ranges showed oedema detectable by clinical observation. They also state that during the first week of acute haemorrhagic nephritis, and during exacerbations of haemorrhagic nephritis caused by injury or infection, transient oedema may recur when the plasma proteins, both total and albumin, are above the critical levels.

COPE (31) found that in "non-oedematous nephritis" the protein osmotic pressure was unreduced, while in "nephritis with oedema" a definite fall in protein osmotic pressure occurs. IVERSEN and NAKAZAWA (71) in "chronic nephritis" without oedema report a total protein content of 7.2 gm. per cent. while in acute nephritis it was 6.12 gm. per cent. CIPRIANI and CIONINI (27) in twelve cases of "acute diffuse glomerulonephritis" with oedema found the total protein to vary between 5.13 and 7.28 gm. per cent., while the colloid osmotic pressure (calculated from GOVAERTS' figures) was 22.8 - 39.8 cm. of water. M'CLURE, De TAKATS and HINMAN (114) in an investigation of oedema of renal type in children report that the total protein and albumin percentage in the plasma was always low when definite oedema was present.

PETERS and his co-workers (119A) in thirty-eight cases of acute nephritis found oedema regularly when protein concentration was below 4 gm. per cent., but it was sometimes present in the early stages of the disease when the protein and albumin were within a pint below normal limits. They also found a "reduction of the serum proteins at the expense of the albumin fraction common in the terminal stages of renal disease".

Ovarian Carcinoma.

These two cases are grouped separately on account of the intrinsic interest of one of them - 0.291. As will be seen from Table VIII the one case had a slight reduction in the plasma proteins, including both fractions, with a consequent fall in the colloid osmotic pressure. The first investigation was made four days before, and the second the day after, paracentesis abdomini had been performed and twelve pints of ascitic fluid had been withdrawn. The second case, however, had the amazingly high plasma globulin figure of 8.22 gm. per cent. This figure was confirmed by a second investigation on the same sample, and when a second sample of blood was examined two days later the globulin content was found to be 9.0 gm. per cent. It is of interest to compare this case with the case of nephrosis reported by SALVESEN (128) where the total protein varied from 8.97 - 10.73 gm. per cent., the albumin varying from 1.69 - 2.56 gm. per cent., and the globulin 7.10 - 8.32 gm. per cent. Another of the few recorded cases with a comparably high globulin figure is the case of myelomatosis mentioned by PETERS and his co-workers (119A) where the total protein varied from 9.8 - 13.1 gm. per cent., the albumin, 2.2 - 2.9 gm. per cent., and the globulin, 7.1 - 10.6 gm. per cent. In this case there was no oedema.

TABLE VIII.

OVARIAN CARCINOMA.

No.	Disease.	Ascites	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
			Per cent.	Per cent.	Per cent.		cm. H <sub>2</sub> O
0.132	Bilateral Malignant Ovarian Disease.	++++	5.755	3.094	2.661	1.163	28.52
	Peritoneal involvement	+	4.725	3.319	1.406	2.361	27.77
0.291	Bilateral Malignant Ovarian Disease.	++++	10.388	2.169	8.219	0.264	32.38
	No Peritoneal involvement.	++++	11.250	2.250	9.000	0.250	34.52
	<u>AVERAGE</u>		8.029	2.708	5.321	1.009	30.80
	<u>MAXIMUM</u>		11.250	3.319	9.000	2.361	34.52
	<u>MINIMUM</u>		4.725	2.169	1.406	0.250	27.77

Analysis of Oedema Fluid.

(i) Subcutaneous Fluid. (Table IX. Figure VIII)

(a) Cardiac:- Nine samples were analysed, the average results being, total protein 0.507 gm. per cent., albumin 0.310 gm. per cent., globulin 0.197 gm. per cent.; colloid osmotic pressure 2.72 cm. of water. The maximum readings were obtained in the only case of subacute bacterial endocarditis in the series, where the total protein content was found to be 0.80 gm. per cent. EPSTEIN (43) found in "cardionephritic" cases that the protein content was 0.100 - 4.62 gm. per cent., while BECKMAN (10) in thirteen cases of cardiac failure found that the protein content was 0.22 - 1.82 gm. per cent. IVERSEN (70) states that the albumin content of oedema fluid is low, and its colloid osmotic pressure seldom exceeds 15-20 cm. of water.

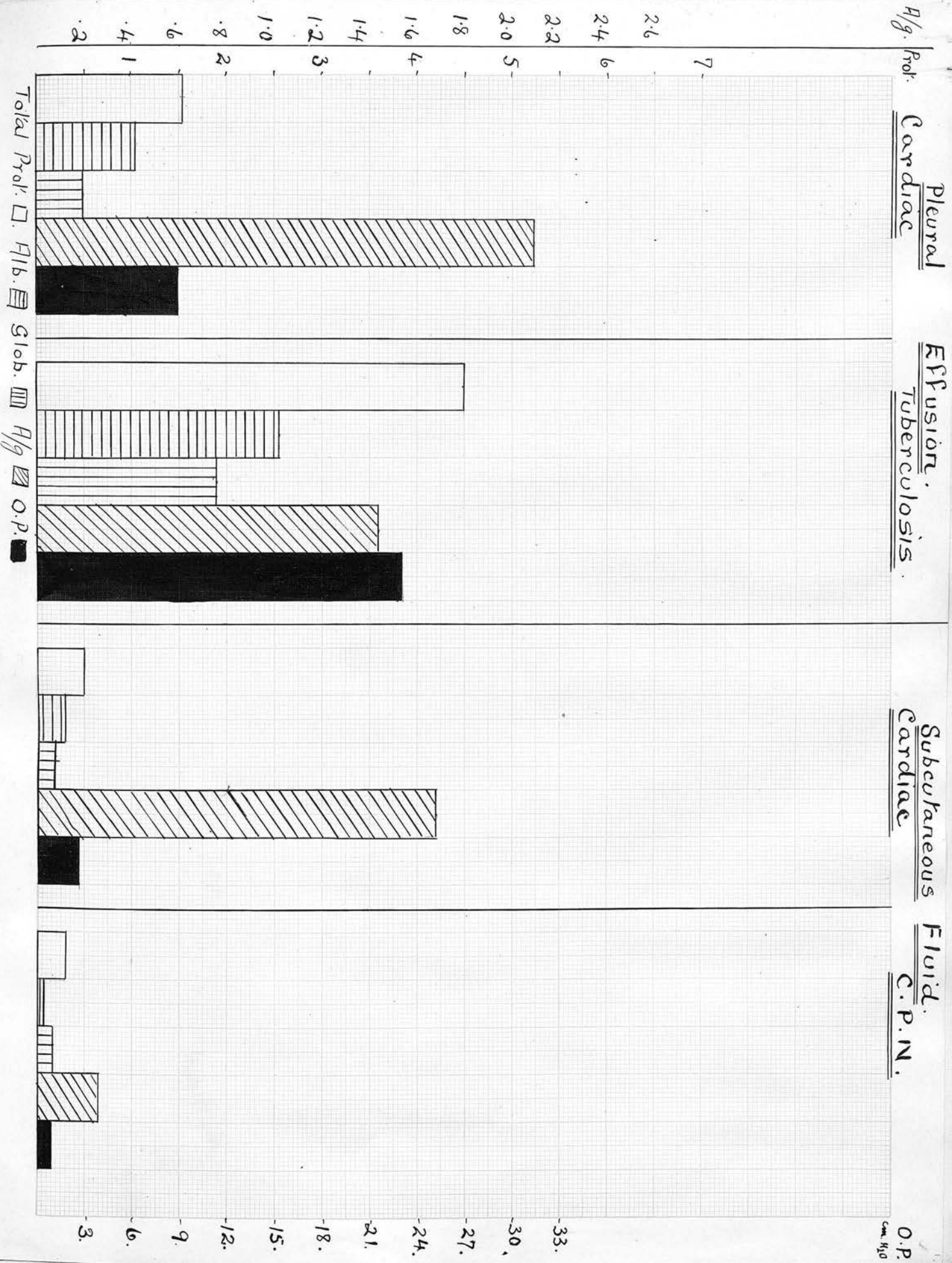
(b) Nephritic:- The one case investigated showed a protein content of 0.276 gm. per cent., the albumin fraction being 0.088 and the globulin 0.188 gm. per cent. with a colloid osmotic pressure of 1.03 cm. of water. EPSTEIN's (43) figures are total protein of 0.098 - 1.171 gm. /



TABLE IX.  
SUBCUTANEOUS FLUID.

No.	Disease.	Amount	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
			<u>Per cent.</u>	<u>Per cent.</u>	<u>Per cent.</u>		<u>cm. H<sub>2</sub>O</u>
0.62	Mitral Stenosis	15 ozs.	0.494	0.369	0.125	2.951	3.03
0.125	Mitral Stenosis) Auricular Fibrillation )	++++	0.350	0.163	0.187	0.872	1.59
0.221	Auricular Fibrillation)	10 c.c.	0.601	0.413	0.188	2.197	3.48
0.98	Myocardial Failure )	35 c.c.	0.263	0.138	0.125	1.104	1.28
0.149	Myocardial Failure )	++++	0.588	0.400	0.188	2.128	3.38
G.83	Mitral Stenosis) Auricular Fibrillation)	23 ozs. 108 "	0.257 0.513	0.163 0.263	0.094 0.250	1.734 1.052	1.41 2.47
0.95	Aortic Incompetence)	Trace	0.700	0.450	0.250	1.800	3.88
0.40	Subacute Bacterial Endocarditis)	++	0.800	0.430	0.370	1.189	3.96
0.108	Chronic Parenchymatous Nephritis )	1 pint	0.276	0.088	0.188	0.468	1.03
<u>Cardiac Oedema.</u>							
AVERAGE			0.507	0.310	0.197	1.679	2.72
MAXIMUM			0.800	0.450	0.370	2.951	3.96
MINIMUM			0.257	0.138	0.094	0.872	1.28
<u>Nephritic Oedema</u>			0.276	0.088	0.188	0.468	1.03

Fig. VIII.



gm. per cent., albumin 0.018 - 0.066 and globulin 0.079 - 0.104 gm. per cent., and in a later paper (44) he gives the average composition of subcutaneous fluid in chronic parenchymatous nephritis as total protein 0.098, albumin 0.080 and globulin 0.018 gm. per cent. BECKMAN (10) found that in four acute and two chronic cases of glomerulonephritis the total protein was 1.12 - 2.52 gm. per cent., while FODER and FISCHER (54) in two cases of acute nephritis found a protein content of 0.39 - 0.96 gm. per cent., while in two cases of "nephrotic glomerulonephritis" the figures were 0.03 - 0.14 gm. per cent.

(ii) Ascitic Fluid. (Table X. Figure IX)

- (a) Cardiac:- In six observations on four cases the average figures were total protein 1.26, albumin 0.77, globulin 0.490 gm. per cent. with a colloid osmotic pressure of 0.76 cm. of water. EPSTEIN (43) in nine cases of cardiac failure found that the total protein varied from 1.567 to 4.712 gm. per cent., in a later publication (44) giving as his average figure, total protein 3.352, albumin 1.199 and globulin 1.788 gm. per cent.

(b) /



TABLE X.

ASCITES.

No.	Disease.	Amount	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
			Per cent.	Per cent.	Per cent.		cm. H <sub>2</sub> O
O. 62	Mitral Stenosis	2 pts.	1.288	0.663	0.625	1.061	6.22
N.186	Mitral Stenosis	7½ "	3.375	1.750	1.625	1.078	16.36
	Cholelithiasis	9 "	0.523	0.283	0.240	1.179	2.60
	Auricular Fibrillation)	7 "	2.690	1.330	1.360	0.978	12.68
M.217	Mitral Incompetence	124 ozs.	1.230	0.790	0.440	1.795	6.81
O.155	Carcinoma of Stomach)	141 ozs.	1.262	0.856	0.406	2.108	7.25
	Arteriosclerosis						
O.153	Tuberculosis	96 "	1.944	1.069	0.875	1.222	9.77
		100 "	1.944	1.131	0.813	1.391	10.11
		174 "	2.138	1.544	0.594	2.600	12.80
O.132	Bilateral Malignant)	12 pts.	3.200	2.700	0.500	5.400	21.33
	Ovarian Disease						
O.291	Bilateral Malignant)	++++	8.625	1.750	6.875	0.255	26.60
	Ovarian Disease.						
	No Peritoneal Involvement.						
	<u>AVERAGE</u>		2.704	1.313	1.391	1.979	12.61
	<u>MAXIMUM</u>		8.625	2.700	6.875	5.400	26.60
	<u>MINIMUM</u>		1.230	0.663	0.406	0.255	6.22
	<u>Cardiac Failure.</u>						
	(3 cases)						
	<u>AVERAGE</u>		1.260	0.770	0.490	1.655	6.76
	<u>MAXIMUM</u>		1.288	0.856	0.625	2.108	7.25
	<u>MINIMUM</u>		1.230	0.663	0.406	1.061	6.22
	<u>Tuberculosis.</u>						
	(3 cases)						
	<u>AVERAGE</u>		2.009	1.248	0.761	1.738	10.89
	<u>MAXIMUM</u>		2.138	1.544	0.875	2.600	12.80
	<u>MINIMUM</u>		1.944	1.069	0.594	1.222	9.77
	<u>Ovarian Carcinoma.</u>						
	(2 cases)						
	<u>AVERAGE</u>		5.912	2.225	3.687	2.827	23.96
	<u>MAXIMUM</u>		8.625	2.700	6.875	5.400	26.60
	<u>MINIMUM</u>		3.200	1.750	0.500	0.255	21.33

- (b) Tubercular:- In three observations on one such case the average figures were total protein 2.009, albumin 1.248 and globulin 0.761 gm. per cent.
- (c) Ovarian Carcinoma:- Two such cases were investigated and as can be seen in Table X, the results show a striking difference. Case 0.291 is the case already referred to (p.23) in which the abnormally high plasma globulin was found, and a further point of interest is that at operation case 0.132, with a moderately low plasma and ascitic globulin, was found to have extensive peritoneal involvement, while case 0.291 showed no suggestion of such at the autopsy. Grouping together ascites due to tuberculosis and new growths EPSTEIN (43) found that the results were, total protein 1.725 - 3.725, albumin 1.444 - 2.219 and globulin 0.281 - 1.838 gm. per cent., while BENNETT, DODDS and ROBERTSON (11) in a case of ? Banti's disease found the ascitic fluid to contain 3.27 gm. per cent. of protein (albumin 1.63 gm. per cent.) and in a case of cirrhosis of the liver a total protein content of 7.7 gm. per cent. (albumin 4.7 gm. per cent.)



(iii) Pleural Fluid. (Table XI. Figure VIII)

- (a) Cardiac:- In five cases the average figures were, total protein 1.571, albumin 1.046 and globulin 0.525 gm. per cent., with a colloid osmotic pressure of 8.91 cm. of water.
- (b) Tubercular:- Three observations were made in two cases, giving an average result of total protein 4.488, albumin 2.571 and globulin 1.917 gm. per cent., the colloid osmotic pressure being 23.13 cm. of water. EPSTEIN's (43) comparable results (three cases) are, total protein 4.275 - 6.250 gm. per cent. (albumin 2.387 - 3.506 gm. per cent.)

It is thus evident that in cardiac oedema the subcutaneous fluid has the smallest protein content, while the pleural fluid has the greatest with ascitic fluid occupying an intermediate position but much closer to the pleural fluid. The protein content is lower in the subcutaneous fluid of nephritic oedema than in that of cardiac origin, though as will be seen later the lowest figures of all are found in nephrotic oedema. In both tubercular and neoplastic "effusions", whether ascitic or pleural, there is a much higher protein content.

TABLE XI.

PLEURAL EFFUSION

No.	Disease.	Amount	Total Protein	Albumin	Globulin	Alb/Glob	Osmotic Pressure
		Ozs.	Per cent.	Per cent.	Per cent.		cm. H <sub>2</sub> O
0.96	Mitral Stenosis) Auricular Fibrillation )	53	0.951	0.513	0.438	1.171	4.72
0.221	Auricular Fibrillation )	15	1.513	1.200	0.313	3.898	9.65
0.230	Myocardial Failure )	20	1.094	0.594	0.500	1.188	5.45
0.98	Myocardial Failure )	32	1.512	1.012	0.500	2.024	8.61
0.162	Arterio-sclerosis )	18	2.788	1.913	0.875	2.186	16.13
0.217	Tubercular Pleurisy with ) Effusion )	18	3.876	2.563	1.313	1.951	21.89
		10	4.388	2.513	1.875	1.340	22.60
0.300	Tubercular Pleurisy with ) Effusion )	2	5.201	2.638	2.563	1.029	24.89
	<u>AVERAGE</u>		2.665	1.618	1.047	1.848	14.24
	<u>MAXIMUM</u>		5.201	2.638	2.563	3.898	24.89
	<u>MINIMUM</u>		0.951	0.513	0.313	1.029	4.72
	<u>Cardiac Failure</u> (5 cases)						
	AVERAGE		1.571	1.046	0.525	2.093	8.91
	MAXIMUM		2.788	1.913	0.875	3.898	16.13
	MINIMUM		0.951	0.513	0.313	1.171	4.72
	<u>Tuberculosis</u> (3 cases)						
	AVERAGE		4.488	2.571	1.917	1.440	23.13
	MAXIMUM		5.201	2.638	2.563	1.951	24.89
	MINIMUM		3.876	2.513	1.313	1.029	21.89

## DISCUSSION.

### THE PLASMA PROTEINS.

Chemistry. The plasma proteins can be divided into four main groups - Fibrinogen, Euglobulin, Pseudo-globulin and Albumin. That these groups, at least the globulins, can be further subdivided is certain, as it is that there are certain other less recognised forms, e.g. Fibrinoglobulin and Seromucoid, but it is not necessary here to go into such full detail.

As to the question of whether there are different albumin fractions in the blood plasma there is considerable divergence of opinion though the following facts summarised by HOWE (69) would seem to offer strong evidence in favour of such an opinion -

(i) Different heats of coagulation and variable distribution of albumin according to the species of the animal; (ii) The separation of fractions having definite limits of precipitation; (iii) The fraction specificity of the albumin fractions; (iv) The variation in albumin fractions under environmental conditions; (v) The impossibility of crystallizing out all the albumin of the serum.

The protein fractions are separated by salting out, the following being the methods of precipitation:-

Euglobulin /

<u>Euglobulin</u>	precipitated by	(1) Saturated solution of Sodium Chloride.
		(2) Half saturated solution of Magnesium Sulphate.
		(3) Third saturated solution of Ammonium Sulphate.
<u>Pseudoglobulin</u>	" "	(1) Saturated solution of Magnesium Sulphate.
		(2) Half saturated solution of Ammonium Sulphate.
<u>Albumin</u>	" "	(1) Saturated solution of Ammonium Sulphate.
		(2) Acidification of saturated solution of Magnesium Sulphate.
		(3) Addition of another salt, such as Sodium Sulphate.

As a means of comparison the following limits for precipitation (in terms of percentages of Ammonium Sulphate) are given:-

Fibrinogen	-	-	30 per cent.
Euglobulin	-	-	30-36 per cent.
Pseudoglobulin I	-	-	36-44 per cent.
Pseudoglobulin II	-	-	44-50 per cent.
Albumin	-	-	50-100 per cent.

Only other two points need be noted in this connection, and they are the most important so far as the relationship of the plasma proteins to oedema is concerned. According to SØRENSEN (137) the molecular weight of serum albumin is 45,000, pseudoglobulin /

pseudoglobulin 80,000 and euglobulin 140,000, which means that a solution of one gramme of albumin contains a much larger number of molecules than a solution of globulin of the same concentration by weight. The other important relevant fact is that the iso-electric point of albumin is further from neutrality than that of globulin, so that at a pH of 7 the albumin is much more dissociated than the globulin. It is the combination of these two factors that accounts for the distinct difference in the osmotic pressure per gramme of albumin and per gramme of globulin.

Origin. "That the protein is of cellular origin is certain; the question is whether or not the fractions arise as identities, with perhaps independent origin, or have a common origin and are inter-related, and finally the immediate source of the proteins. The 'life-cycle' of the proteins is equally obscure". (HOWE, 69). In the chicken embryo the bloodvessels arise from cells called angioblasts which differentiate from the mesoderm and produce endothelium, blood plasma and red corpuscles. In other words the first blood plasma results from the destruction of cells and is different /



different in origin from tissue fluid. In the adult, however, according to SABIN (126), the origin is not from the endothelium of bloodvessels, though it is possible that the endothelial liver tissue continues to function in this way. A further possibility is that the periodic disintegration of the neutrophil leucocytes may be the source of at least part of the blood proteins.

KERR, HURWITZ and WHIPPLE (73) have shown that serum proteins can be regenerated when there is no protein intake in the food, the deduction being that under such circumstances the serum proteins must be formed from body proteins. Their experiments also give no evidence that the serum proteins may be in any way concerned as intermediary products between food protein and body tissue or parenchyma protein, and they also showed that the regeneration of the serum proteins is more rapid and complete upon a meat or mixed diet than upon a bread and milk diet. In addition they make the suggestion that blood protein construction may depend upon the activity of all protoplasm, it being possible that all protein material may be built up part way alike, to be differentiated in the last stage of the complete elaboration. Finally they report the interesting parallel /

parallel that the regeneration of the serum proteins, after fifty per cent. depletion, on a meat diet may be effected in five to seven days and that the regeneration of liver cell protoplasm, after fifty per cent. necrosis, may be effected in the same period. As further evidence in favour of a close connection between the liver and the blood proteins they found that pronounced injury of the liver by means of phosphorus or chloroform caused, or at least was associated with, a moderate fall in blood serum protein, while regeneration of the blood serum proteins following a plasma depletion is delayed by the presence of liver injury due to phosphorus or chloroform.

SMITH, BELT and WHIPPLE (135) using a slightly different technique, found that after rapid blood plasma depletion, the increase in serum protein concentration begins at once, is very rapid during the first fifteen minutes and more gradual thereafter. They accordingly suggest that this indicates some reserve supply, perhaps held in the body cells, this emergency reserve evidently being small and the production of further supplies being difficult and requiring time. The fibrinogen reacted differently, the same initial fall not being followed by a rapid rise in the first quarter of an hour, but recovery being /

being complete in twenty hours, as compared with two to seven days in the case of the albumin and globulin fractions.

SEITZ (133) was also of the opinion that the liver acted as a storehouse for the blood proteins, and that after any loss these were restored from this reserve. MORAWITZ (112) and also HIROTA (68) however suggested that following severe haemorrhage the protein from the body tissue fluid may enter the capillaries directly, a suggestion which FIELD and DRINKER (49) tend to support. BARKER (6) has reported that dogs on a diet theoretically adequate in calories, minerals and vitamins, but deficient in nitrogen, gradually developed fatty degeneration changes in the liver. This fatty replacement became extensive if the diet were continued but if protein were gradually added the animal would usually recover.

Thus, in spite of the absence of any definite knowledge, there would seem to be very sound reasons for believing that the liver, if not the site of formation of the blood proteins, is at least closely associated with their formation, and that the fractions arise as separate identities.

Species. In man, sheep, goat, rabbit, dog, guinea pig and rat, albumin predominates over globulin, while in the horse, hog and cow the two fractions are equal or the globulin predominates.

Age. There is a distinct variation with age. LEVY (93) in a series of twenty-nine infants found the serum proteins to vary from 5.6 gm. per cent. in the first month of life to 7.5 gm. per cent. by the end of the first year. In a series of young dogs CLARK and HOLLING (28) found that the rate of increase of the blood proteins was rapid up to the fortieth day, less rapid thereafter, attaining the adult level about the sixtieth to seventieth day. It is also of interest that their animals were all weaned at the end of the fourth week and before this all points lay on a curve, while after this they became more scattered.

Sex. According to HOWE (69) sex differences have not been demonstrated, but during pregnancy there is an increase in the total protein of the maternal blood due to a rise in the globulin protein. OBERST and PLASS, on the other hand, give the following average figures (116):-

Non-pregnant /



	<u>Total Protein</u> <u>Gramme per cent.</u>	<u>Cases.</u>
Non-pregnant women	7.22	16
Early Pregnancy (1-3 months)	6.92	10
Late Pregnancy (8-9 months)	6.43	33
Parturient women	6.44	25
" " (7-9 days)	7.16	14

Function. Fibrinogen of course has the unique function among the plasma proteins in relation to the stoppage of haemorrhage, but apart from this the two main functions of the blood proteins are as stabilising factors and in relation to the control of water movement in the body. This latter function will be discussed in the next section. With regard to the former SMITH, BELT and WHIPPLE (135) showed that rapid depletion of the blood protein can rarely be carried below one gramme per cent. without causing a fatal reaction. In a subsequent paper (152) they showed that when the Locke's solution in the red cell mixture used in plasmapheresis was replaced by serum the toxic effect of plasma depletion was completely removed. "This gives a strong indication that the blood serum proteins are stabilising or protective factors." They are the essential environmental factors of the circulating blood in its relation to the body cells, and this may be the most important function of the plasma colloids.

### FACTORS CONTROLLING OEDEMA FORMATION.

In his monograph on "Oedema", LOEB (99) under the heading of "Multiplicity of Factors in Oedema", summarises the situation by stating that there are three sets of factors which regulate the normal movement of water and dissolved substances -

(i) Primary factors which represent a more or less connected mechanism which includes absorption, circulation and elimination of fluid; (ii) Secondary factors - diffusion and osmotic potentials and perhaps endoelectrosmotic forces, changes in the permeability of membranes, and probably, to a limited extent, filtration pressure; (iii) The means through which these "secondary factors" exert this effect -

(a) distribution of sodium chloride and proteins, both of which <sup>are</sup> of special importance in establishing and regulating osmotic and diffusion potentials, and protein aggregates may retain water in a specific manner; (b) factors which regulate the permeability of membranes, e.g. a certain equilibrium of ions, the action of certain hormones.

It is not the purpose of this paper to enter into a full discussion of all these primary factors - that is a subject which requires a full and independent survey - but it is proposed to deal with the more important /

important factors in the causation of oedema, and particularly those which have a bearing on the relationship between the plasma proteins and oedema.

#### Capillary Blood Pressure.

One of the obstacles in the way of a clear understanding of cardiac oedema is that no satisfactory method has yet been evolved of measuring the capillary blood pressure in man. In the past three main methods have been used - (i) The pressure required to cause a certain degree of blanching of the skin; (ii) Microscopic observation; (iii) Direct measurement of the pressure by puncturing the capillary.

The older estimations are so variable as to be of little value, but of the more recent three may be quoted. DANZER and HOOKER (32) using a method depending on the production of stagnation of fluid of the corpuscles in the capillaries, found the normal capillary blood pressure in the sitting posture to be 22.2 mm. of mercury, while ELLIS and WEISS (39) give 9 mm. of mercury as being the average in normals. The latter used a colour blanching method. LANDIS by means of direct measurements in the human skin found the average capillary pressure in the arteriolar /

arteriolar loop to be 43 cm. of water, and in the venous loop 16 cm. of water.

It is in relation to cardiac oedema that the question of the capillary blood pressure is most relevant, and while there is general agreement that a rise does occur the exact importance of it is variously assessed. STARLING (139), for instance, gives increased intracapillary pressure as one of the factors involved in the causation of dropsy. He states that if in a normal animal venous obstruction occurs, the first effect is a rise of capillary pressure, but this in itself is not sufficient to cause oedema, and again in the same paper, discussing the oedema of uncompensated heart disease he states that in the first stage there is a fall of capillary pressure in the peripheral parts of the body, in the kidneys and in the intestines, while in the third stage there is a rise of capillary pressure in all dependent parts. In a subsequent paper (140) on the pathology of heart disease he shows that if a limb is enclosed in a plethysmograph and oil is then injected into the pericardial sac, there is no change in the volume of the limb while the arterial pressure is constant, even though there may be a rise of pressure in the portal vein and vena cava, but as /



as soon as the arterial pressure falls, the volume of the limb diminishes. He then goes on "We may conclude therefore that even with a coincident vasoconstriction the effect of heart failure must be a fall and not a rise of blood pressure in the capillaries and smaller veins of the limbs". He also quotes COHNHEIM (29) as being of the opinion that in heart disease the capillary pressure is not higher, but rather lower, than normal.

One of the earliest reported experiments is that of LOWER (100) who ligatured the inferior vena cava in the chest in animals and obtained ascites. He also ligatured the jugular veins at their lower ends and obtained oedema of the head and neck, concluding that the cardiac oedema was due to an increase of pressure in the veins and capillaries.

BOLTON (16), as a result of the experimental production of uncompensated heart disease by constricting the pericardium by sutures, is of the opinion that it is entirely a matter of speculation whether the capillary pressure rises, falls or remains normal when the venous pressure is raised and the arterial lowered, the essential point being that when the venous pressure has fallen to normal, the arterial pressure remaining low, the capillary pressure /

pressure must be low. In the experiments he found that the high venous pressure which occurred was only temporary and even followed by normal venous pressure. In a subsequent series of experiments (17), in which he produced dropsy by ligaturing the inferior vena cava above the diaphragm, he concludes that increased venous pressure, the immediate result of venous obstruction, leads to dilatation of the bloodvessels behind the obstruction and filling out of the capillaries, and so is an indirect factor. It is not a direct cause, because oedema occurs when the pressure is normal. When dropsy is produced with a high capillary pressure, such a pressure becomes an important contributing factor. His final conclusion is that the capillary pressure varies at different stages of the disease and in different parts of the body.

LEONARD HILL (65) also tends to depreciate the role of capillary pressure in oedema - "I ask you to consider not increased capillary pressure and filtration as the causes of oedema, but stagnation of fluid with consequent oxygen want and increased imbibition".

DAUTREBANDE, DAVIES and MEAKINS (34) in reviewing /

reviewing their experiments, in which extreme stasis of the arm was produced, state that the occurrence of gross oedema could not with certainty be observed. Such swelling as occurred during the application of stasis was mainly of the nature of plethora. In such circumstances the increase of venous and capillary pressure must have been considerable, and hence the absence of gross oedema in itself is an argument against the mechanical pressure theory.

MELDOLESI (108) on the other hand has reported a rise in the capillary pressure during the accumulation of oedema.

The position would thus seem to be that, though there may be, and indeed probably is, a rise in the capillary blood pressure in cardiac oedema, it is by no means one of the prime factors in the production of oedema.

#### The Permeability of the Capillary Wall.

It is known that the capillary walls are freely permeable to the crystalloids and salts of the blood, but there is still a considerable difference of opinion as to whether they are normally permeable to the plasma proteins, and as it is this latter factor which is of the greater interest from the /

the point of view of oedema formation, it is proposed to devote some time to its consideration.

CHURCHILL, NAKAZAWA and DRINKER (26) after demonstrating that the protein content of normal frog lymph varies from 0.29 to 2.17 gm. per cent., infer a degree of permeability to protein on the part of the walls of adjacent capillaries which is entirely unlike that "commonly assumed to exist in mammals except in certain tissues such as the liver". Similar deductions were made by CONKLIN (30) in a study of the formation and circulation of lymph in the frog, the conclusion arrived at being that the capillaries of the frog's skin are normally permeable to protein. In a series of papers FIELD and DRINKER (48) came to the conclusion that the capillaries of the dog are permeable to protein and that "lymph is tissue fluid". These conclusions they base on their finding that in the dog lymph invariably contains albumin, globulin and fibrinogen. They suggest that the capillaries practically universally leak protein, that the filtrate from the capillaries to the tissue spaces contains water, salts and sugar in the concentration found in the blood, together with serum albumin, globulin, and fibrinogen in low concentrations, probably lower than /



than that of lymph or tissue fluid, that water and salts are absorbed by the bloodvessels, while the protein enters the lymphatics together with water and salts in the concentration existing in the tissue fluid at the moment of entrance into the lymphatics.

(35A). As a result of experiments on dogs with infiltration of horse serum they conclude that the capillary endothelium is permeable to protein in both directions, although, under normal pressure and osmotic relations, the protein which has left the blood is returned by the lymphatic route alone.

(49) In another series of experiments they showed that changes in the protein content of the blood, whether due to an injured capillary wall or to a great reduction in the circulating blood proteins, were almost immediately reflected in alterations in the protein concentration of the lymph. "These changes can only mean that the capillaries of the dog are partially permeable to protein molecules, and that, along with the passage of diffusible contents, there is also the passage of the less diffusible colloids". (50)

LANDIS, on the other hand, is of the opinion that the capillary wall is normally impermeable to protein, and suggests as an explanation of the discrepancy /

discrepancy between his results and those of other workers, that the tissue fluid immediately outside the capillary walls can differ widely in composition from the lymph even of the same area.(85). In a subsequent paper, with KROGH and TURNER (80) he concludes "From the data available at present, it seems most likely that the normal capillary wall is not absolutely impermeable to protein and that an exceedingly slight leak of protein occurs rather generally. The concentration of protein in the capillary filtrate must certainly be less than 0.1 per cent., and the relatively high protein content of lymph is probably due to reabsorption of water and salts from the original capillary filtrate". Finally he shows that caution must be used in extending conclusions based on observations involving high grades of venous congestion to considerations of normal capillary permeability.

From a survey of the available work it would seem that this latter conclusion is the correct one - that the normal capillary wall is very slightly permeable to protein, a conclusion the significance of which will be referred to again.

With regard to cardiac oedema, there is a fairly general agreement that there is an increased permeability /

permeability of the capillary wall. LAZARUS-BARLOW (89) while certain that malnutrition of the tissues and accumulation of waste products were the primary factors in the production of dropsy, admitted also that the vessel walls were altered. COHNHEIM (29) on the other hand considers increased capillary permeability one of the primary factors.

STARLING (139) is of the opinion that, in cardiac oedema, as a result of the failing circulation there is stagnation of the blood in the bloodvessels, and the oxygenation and nutrition of the capillary walls cannot be normally carried out, so that their permeability is increased. BOLTON (17) goes so far as to say that it is probable that passive oedema is primarily due to impairment or other alteration in the capillary wall, and in a subsequent paper - "In the normal condition it seems unlikely that such a membrane as the capillary wall should be absolutely independent of physical laws; and, on the other hand, it appears that, being a lining and actively metabolic tissue, it does not allow substances to pass through it as though a dead membrane. In the diseased conditions of venous obstruction, therefore, the normal activity of the capillary endothelium is overshadowed by a passive /

passive transudation through the damaged membrane."

LOEB (99) in his critical survey of the whole subject of oedema concludes from the evidence then available that dilatation of the capillaries and veins due to faulty functioning of the circulation is the principal cause of increased transudation, and subsequent oedema in cardiac decompensation. This conclusion, however, is reached with considerable reluctance, as in a subsequent part of his survey he expresses the opinion that the evidence we have in the majority of cases of clinical oedema as to the insignificance of changes in the permeability of vessels is either circumstantial or even contradictory. "In cardiac decompensation we assume that the dilatation of the capillaries combined with an increase in carbon dioxide tension in the capillary blood may increase permeability".

Since then, however, a considerable amount of experimental work has been done, particularly by LANDIS, which has given us definite data on this subject. KROGH (79) had already expressed the opinion that the increased permeability after the application of urethane was entirely due to dilatation of the capillaries. LANDIS (83) on the other hand by his micro-injection method showed in the /



the frog that increased permeability to urethane occurred in contracted as well as dilated capillaries, and that the primary cause of the increased permeability after urethane is the toxic effect of the drug in the capillary wall. Subsequently he showed, again in the frog, that the rate of passage of a dye solution through the capillary wall appeared to depend upon the level of capillary pressure, not upon the capillary diameter (84).

His most vital experiments (85) were those in which he showed that, immediately after a three minutes' period of oxygen lack, fluid filtered through the capillary wall at approximately four times the normal rate, and that the increased permeability of the wall also permitted the passage of protein, thus reducing the effective osmotic pressure of the plasma proteins, indicating that the wall still acted merely as a passive filter, though more permeable than normal. He also showed that exposure of the mesentery to Ringer's fluid half saturated with carbon dioxide produced no change in fluid movement, while complete saturation only increased the rate of fluid movement very slightly, the wall remaining normally impermeable to protein. Similarly it was demonstrated that an increase in hydrogen /

hydrogen ion concentration produced almost no change in fluid movement within physiological limits, although at pH 4.0 the characteristic effects of injury appeared. From these experiments he draws the conclusion that they indicate the probable importance of oxygen lack as a factor in producing the changes in fluid movement observed during cardiac decompensation. In regions with a sluggish blood flow oxygen lack might well produce permeability changes, particularly on the venous side of the capillary network, or in capillaries in which the intermittency of flow was unduly prolonged.

In experiments on the human capillary wall, carried out in conjunction with JONAS, ANGEVINE and ERB (87), he showed a straight line relationship between venous pressure and filtration rate, when the latter is measured by the plethysmograph during venous congestion of 11-39 mm. of mercury, and with the highest venous congestion produced (80 mm. of mercury) there appeared to be a measurable loss of protein through the capillary wall. By what mechanism this protein escapes they are not prepared to say, although the presence of red cells in the lymph and of the petechiae frequently noted in the skin suggest a mechanical rupture of the capillary wall. /

wall. The fact that the lymph protein in general decreases during such congestion, in spite of the presence of red cells, does not eliminate a mechanical rupture, since the cells and plasma escaping through a few weak spaces in the wall may be diluted by the relatively protein-free filtrate from the remainder of the capillary.

This survey of the literature suggests strongly that in the oedema of cardiac decompensation there is an increased permeability of the capillary wall.

Brief reference may be made to other conditions in which such an increase in permeability occurs. In acute nephritis it has been known that the correlation between the level of the plasma proteins and the onset of oedema is not so accurate as in other forms of nephritis. PETERS and his co-workers (118) have suggested that this anomaly may be due to a toxic effect on the capillary walls resulting in an increased permeability. Again, in inflammatory oedemas the outpouring of fluid is associated with increased capillary permeability, as has been demonstrated by, among others, MEAKIN and MEAKIN(107) who showed that the rapid accumulation of dye in an inflamed area was associated with increased capillary permeability.

"Kolloidosmotischen".

It was STARLING (138) who, in what must now be considered an historic publication, first drew attention to the importance of the fact that, although the osmotic pressure of the plasma proteins is so insignificant, yet it is of an order of magnitude comparable to that of the capillary pressure; and whereas capillary pressure determines transudation, the osmotic pressure of the serum proteins determines absorption. Thus at any given time there must be a balance between the hydrostatic pressure of the blood in the capillaries and the osmotic attraction of the blood for the surrounding fluids.

As to the exact origin of the osmotic pressure of the plasma proteins there is still some difference of opinion. SORENSON (136), after a study of the physical properties of a solution of crystallised egg albumen in the presence of sodium sulphate, came to the conclusion that the osmotic pressure of a solution of egg white is a very definite quantity which depends not only on the concentration of the protein but also on the concentration of the solution in ammonium sulphate and hydrogen ions; yet for a solution of known composition the osmotic pressure, within /





within the limits of experimental error, always remains the same.

LOEB (98) has shown that proteins can be considered as amphoteric electrolytes which, as he had previously shown (97) are capable of forming salts either with alkalies or acids, depending upon the hydrogen ion concentration. On the acid side of the isoelectric point ( $\text{pH} = 4.8$ ) they can only combine with the anion of neutral salts, on the less acid side of the isoelectric point only with cations, while at the isoelectric point either with cations or anions of a neutral salt. At pHs other than the isoelectric point proteins exist as salts of proteins or alkaline proteinates and are practically dissociated into a protein ion and a positive or negative ion. The true osmotic pressure of proteins can only be measured at the isoelectric point when it is very small. The pressure attained in the osmometer by a solution of gelatin or albumin is the osmotic pressure due to the presence of colloids, but which really results from the unequal division of crystalloids between the two sides of the membrane. This inequality of division of ions is known as the DONNAN Equilibrium, which has been summarised by FREUNDLICH (55) as follows:-

In /



In a solution of congo red, the anion is colloidal and does not diffuse through a colloidal membrane, while the Na cation passes through. Now if an osmotic cell contains congo red and another salt, e.g. sodium chloride, having the same cation, this salt does not distribute itself uniformly between the liquids inside and outside the cell and quite independently of the congo red. When the concentration of sodium chloride is small, congo red hinders the entrance of the salt into the cell and more of it remains outside the cell than inside it. If the congo red solution is dialysed against pure water, the outside liquid becomes alkaline, while the dye solution becomes darker and turbid, as a result of the formation of the dye acid. This depends on the fact that the Na ion passes through the membrane; that of congo red does not. In order that the Na ions may have an equivalent amount of anions hydrolysis occurs; OH ions pass through the membrane, while an equivalent quantity of H ions are found in the dye solution.

RUSZNYAK (125) also attributed the measurable osmotic pressure of the plasma proteins to the DONNAN Equilibrium and emphasised the fact that at the ordinary pH of the plasma, the albumin fraction is /

is farther removed from its isoelectric point than the globulin fraction, and so had a higher osmotic effect per gramme of protein.

HASTINGS and his co-workers (61), on the other hand, showed that the sum of the osmotic pressures of isoelectric albumin and globulin, calculated from the known molecular weights and their concentration in normal serum, were more than five times the pressure due to the DONNAN Equilibrium. Similar deductions were drawn by MARRACK and HEWITT (103) who calculated on theoretical grounds that under normal conditions a change in pH from 7.0 to 8.0 should give rise to an increase in osmotic pressure of 6.5 cm. of water on the basis of the DONNAN Equilibrium, whereas actual measurements gave a rise of not more than 1.0 cm. of water for a change in pH of 6.8 to 8.0. Neither view is incompatible with the other and the position is perhaps best expressed by FAHR and SWANSON (45) when they refer to the "effective" osmotic pressure of the plasma proteins, this being "the osmotic pressure actually exerted by these proteins minus the counter pressure due to the DONNAN Equilibrium".

A further point to which reference may be made is that VERNEY (148) and SCHADE and CLAUSEN (129) both /

both expressed the opinion that the relation between the osmotic pressure of the serum proteins and the percentage concentration of these proteins was not a linear one, VERNEY, for instance, stating that dilution of the plasma with Ringer's fluid gives rise to a relatively larger fall in the osmotic pressure than the concomitant fall in protein concentration. VON FARKAS (46) on the other hand, found that if the serum was moderately diluted (3-7 gm. per cent. of protein), the osmotic pressure remained parallel to the concentration of the total proteins. This latter finding suggests that what variation there may be at higher dilutions is of comparatively little value clinically.

GOVAERTS (57) has shown that between the albumin-globulin ratio and the osmotic pressure per gramme of protein there seems to exist a definite persistent relationship, and from his results he has calculated, with only a ten per cent. error, that the osmotic pressure per gramme of albumin is 7.54 cm. of water, while the osmotic pressure per gramme of globulin is 1.95 cm. of water. These results he has subsequently confirmed (58) in a large series of cases and has shown that the figures based on these figures correspond very closely with those obtained /



obtained by actual measurements. A further confirmation is to be found in the work of VON FARKAS (46) who, using SCHADE's onkometer, estimated the osmotic pressure per gramme of albumin to be 6.8 cm. of water, and per gramme of globulin 2.51 cm. of water. The resemblance of these figures to those of GOVAERTS is very significant, particularly as different apparatus was used by the two workers. The difference in absolute value results from the fact that with SCHADE's onkometer a smaller osmotic pressure is obtained for normals than with GOVAERTS'.

Having thus established that the plasma proteins do exert a definite osmotic pressure it is now necessary to consider briefly the part played by this pressure in the normal fluid exchanges of the body. CUSHNY (31A) was of the opinion that filtration through the glomeruli was only possible if the hydrostatic pressure in the capillaries was greater than the osmotic pressure of the plasma proteins or, what the Germans have termed, the onkotic pressure of the blood. This has been confirmed by various workers including WHITE (153) who showed that in the frog the onkotic pressure was of such a magnitude as to be exceeded by the pressure in the glomerular capillaries. On three frogs /

*John White 153*

TABLE XII.

RELATION between CAPILLARY BLOOD PRESSURE and  
OSMOTIC PRESSURE of PLASMA PROTEINS.

	Capillary Pressure	O.P. of Plasma Proteins	Venous Capillary Pressure
	<u>cm. H<sub>2</sub>O</u>	<u>cm. H<sub>2</sub>O</u>	<u>cm. H<sub>2</sub>O</u>
RAT	30.0	25.5	17.0
		25.5	
		22.0	
		25.0	
		25.0	
		26.5	
GUINEA PIG	38.5	22.0	17.0
		27.5	
		24.0	
FROG	14.5	11.5 (Landis) 10.0-11.9 (White)	10.0

frogs he obtained the following results:-

	<u>Osmotic Pressure</u> <u>of Colloids.</u> <u>cm. plasma.</u>	<u>Percentage</u> <u>Plasma Proteins.</u>
(1)	9.6	2.40
(2)	9.8	2.52
(3)	11.5	2.80

while the capillary pressure was 14.5 cm. of water

SCOTT (132) came to the conclusion that every fall of blood pressure resulted in an inflow of fluid into the bloodvessels, while every rise of blood pressure resulted in an outflow of fluid. Similarly ZUNG and GOVAERTS (158) showed that dogs with low plasma proteins withstood large haemorrhages particularly badly.

This relationship was finally established experimentally by LANDIS (86) in the paper from which Table XII is taken. As can be seen from this table in mammals, as well as frogs, at the arteriolar end of the capillary the hydrostatic pressure exceeds the onkotic pressure, while at the venous end it is less. In other words at the arteriolar end of the capillary fluid passes out from the blood into the tissues, while at the venous end the reverse holds good - fluid passing from the tissues into the bloodstream. In this there is a regular and continuous /

continuous fluid interchange between the blood and the tissues of the body. LANDIS had already found in the frog by direct measurement that the rate of movement through the capillary wall was directly proportional to the difference between the capillary pressure and the osmotic pressure of the plasma proteins, amounting to approximately 0.03 cubic micron per square micron of capillary wall per second.

It is of relevant interest to note that WELLS (150) from the study of the concentration and osmotic pressure of the proteins of intestinal lymph from the mesenteric ~~lacteals~~ of dogs as well as of the blood serum, comes to the conclusion that "the protein content of the lymph is determined within fairly definite limits by the protein content of the serum; . . . and that the mesenteric venous pressure is sufficiently high, in relation to the calculated "effective" osmotic pressure, to account for the continuous formation of intestinal lymph by a process of rapid filtration of protein-containing fluid through the capillaries". And again "The findings indicate that the absorbing force of the intestine is due to the osmotic pressure, exerted against the semi-permeable epithelial /



epithelial membrane of the intestine, of the fraction of total serum proteins to which, on the average, the walls of the blood capillaries of the villus are permeable". (151)

It is thus evident that the plasma proteins, by means of their osmotic pressure, play a very considerable part in the fluid exchanges of the blood, and it only remains now, before turning to a survey of the main clinical types of oedema, to summarise briefly the experimental evidence which suggests the essential relationship between hypo-proteinaemia and oedema. This experimental work is largely based on the process which ABEL, ROWNTREE and TURNER (1) named "plasmapheresis" and which they described as "quantities of blood plasma may be withdrawn from an animal without apparent injury that exceed several times the maximum quantity of blood that can be safely drawn by the usual method of venesection, provided that the corpuscular elements of the blood suspended in Locke's solution (0.6 per cent. sodium chloride) be returned to the vascular system after each bleeding."

LEITER (90) demonstrated in dogs that oedema was always produced by plasmapheresis when the plasma proteins were maintained at or below 3 gm. per cent. In these experiments the oedema fluid had a protein content /

content of less than 0.25 gm. per cent., and in his confirmatory experiments (91) he states that cardiac damage, starvation and the alkalinity of the Locke's solution had all been controlled. BARKER and KIRK (7) in a much smaller series of dogs and with a much slower plasmapheresis, obtained a gradual decrease in the serum proteins, particularly the albumin fraction, and whenever the albumin fell below 0.8 gm. per cent. oedema appeared. DARROW, HOPPER and CARY (33), also working on dogs, found the critical level for the total proteins to be 3-3.5 gm. per cent., while for albumin it was 1.5 gm. per cent. Similar results are reported by LEPORE (92) who, however, tends to minimise the importance of the hypoproteinaemia, and to refer to the oedema thus produced as a sodium chloride oedema. He does not claim, however, that sodium chloride alone would have produced the oedema, and it is evident from his protocols that plasma protein depletion was required in each case before oedema began to occur, so that there would seem to be little doubt that the hypoproteinaemia was the primary factor and the sodium chloride content a secondary, though important, factor.

The results of BARNETT, JONES and COHN (8) are of special interest as they emphasise a point which is /

is probably of considerable significance in the aetiology of clinical oedema. Removing only small amounts of blood - 25-100 c.c. - daily they found no significant drop in the plasma protein concentration and no oedema. As a result of their experiments they suggest that the loss of considerable amounts of protein is not alone sufficient to produce lowering of the level of the plasma proteins and that in nephritis of the degenerative type there may be an associated interference with the mechanism of regeneration.

Whatever may be the exact relationship between loss of protein and plasma protein deficiency, there has for long been considerable divergence of opinion as to the relationship between the urinary protein and the plasma protein. ANDREWS and THOMAS (4) using precipitin tests in cases of experimental uraemia and ether nephritides in dogs, claimed that in the early stages urinary proteins originated in the liver and that small amounts of spleen protein had also been recognised, while in the later stages serum protein was present. Their explanation was either (a) in nephritis cellular disintegration occurred to such an extent that protein leaked from the great parenchymatous organs into the blood and was /

was excreted, and that the prolonged passage of foreign proteins through the kidneys rendered them permeable to the blood proteins; or (b) that the serum proteins passing into the tissues underwent "some slight change in structure or acid formation which transformed them just enough to render them susceptible of excretion by the kidneys as foreign products". This view was subsequently modified slightly by THOMAS (145) who, adopting the second alternative mentioned, contended that when the tissue protein has been split to a toxic stage, it is combined with the serum proteins, which combination constitutes a foreign protein and is excreted. "The presence of serum protein in the urine in nephritis is a detoxicating mechanism".

This view has never been confirmed and is opposed by the findings of most workers. HEWITT (64) for instance, came to the conclusion that "no abnormal liver albumin occurs in the urine of nephrosis, albuminuria of pregnancy and eclampsia, and the globulin fraction where liver proteins might occur, is generally very small". EASTMAN (38) differs from HEWITT in finding an extremely high globulin content in the urine in eclampsia, and suggests that this is the direct result of the increased /



increased capillary permeability associated with this disease. The whole problem has since then been surveyed by KERRIDGE and BAYLISS (74) whose final conclusion is that "whether or not a protein is excreted by the kidney depends on its physical properties and not on its chemical nature or biological origin". In a subsequent paper (9) they came to the conclusion that their results are "explained by the filtration theory of glomerular filtration if it be assumed that the glomerular membrane is permeable not only to crystalloids but also to proteins of a molecular weight less than 70,000." They also found that on histopathological examination there was no evidence of renal damage in the dogs which had been injected with protein. CAVETT and GIBSON (25) have also shown that the racemisation curves given by albumins and pseudoglobulins from nephritic urines were similar to those of the corresponding serum proteins. Incidentally they also found that the pseudoglobulin from the blood of a nephritic patient gave a curve identical to that of normal blood.

### THEORIES OF OEDEMA FORMATION.

Two general theories of oedema formation must now be referred to briefly. FISCHER (51,52), as a result of his work on colloids and turgescence, advanced the theory that in the oedema of renal and cardiac origin, the connective tissue becomes acid, and that its increased H-ion concentration produces turgescence resulting in oedema. Unfortunately for the validity of the theory it was shown that in these diseases the connective tissue does not become acid, and also that in this tissue an increased H-ion concentration leads to deturgescence and not turgescence.

ELWYN (40) has elaborated a theory of oedema formation which is based on a regulation of water exchange consisting of:-

- (a) A regulating centre in the interbrain closely connected with the centres for the regulation of sodium chloride metabolism, other mineral exchanges, protein, carbohydrate and fat metabolism and other vegetative centres.
- (b) Reservoir organs which take up any excess of water from the blood, e.g. the muscles and skin, especially their connective tissue, the liver and possibly also the spleen.
- (c) /

- (c) Barriers to the blood-stream consisting of membranes in the tissues and the walls of capillaries through which the stream of water is regulated.
- (d) Excretory organs, chief of which are the kidneys, but in addition the lungs, the skin and the intestines, through which water is eliminated

With this as his basis he assumes that in oedema a change in the electrolytes responsible for the slowing in the rate of water movement is produced reflexly by way of the central nervous control acting through the vegetative nerves or through hormonal influences, or in both ways. In such a system, top-heavy with vegetative centres and based entirely on assumption, it is perhaps only natural that the way to a clearer understanding of oedema seems rather more remote than usual.

CLINICAL /

## CLINICAL TYPES OF OEDEMA.

### Nutritional Oedema.

There is no more intriguing chapter in the history of medicine than that dealing with the dropsy which has devastated besieged cities or stricken armies. The mere variety of names under which it has masqueraded throughout the centuries is proof of its prevalence - war oedema, nutritional oedema, prison dropsy, epidemic dropsy, hunger swelling etc. etc.

Reference has already been made to what must be one of the earliest recorded cases - Heraclitus - and since then many references are found to it in historical records. It was for instance rife in the Peloponnesian Wars (430-425 B.C.)(122); the French soldiers during the siege of Naples in 1528 were seen "with pallid visages, swollen legs and bloated bellies" (62); during the siege of Paris in 1870-71 the inhabitants of the doomed city, particularly the children, suffered from dropsy (146); it was rampant in the concentration camps during the Boer War and in the Russo-Japanese War (104). The list could be extended indefinitely, but it is only in so far as it casts any light on the relationship between plasma protein deficiency and oedema that we are concerned with it here.

It /



It was the recurrence of this dropsy in Central Europe during the last European War that refocussed attention on this subject, and led to the matter being fully investigated. The first cases appeared in prison camps in July 1915, and from then onwards the condition became increasingly common in Germany (MAASE and ZONDEK, (102)), Poland (BUDZYNSKI and CHELCHOWSKI, (23)) Italy (BOLAFFIO (15)) and generally throughout Europe (GUILLERMIN and GUYOT (59)). It was generally realised that the causative factor was a diet low in protein and containing a large amount of fluid and salt, potato being one of the main constituents of the diet. There was considerable divergence of opinion, however, as to the precise cause of the oedema, various factors being incriminated, such as endocrine disturbance increased capillary permeability, excess fluid and salt intake, vitamin deficiency etc. BIGLAND (12), for instance, in discussing oedema as a symptom of "so-called food-deficiency diseases", concludes that "whatever the true explanation may be, it is extremely probable that food deficiency, together with disordered function of certain endocrine organs, plays a large part in the causation of epidemic dropsy and pellagra."

JENSEN (72) and SCHITTENHELM and SCHLECHT (131) had already reported that in their cases the blood proteins /

proteins were nearly always decreased, 4.0-6.4 gm. per cent., and NIXON (115) again drew attention to this factor, giving as the causes of "famine-dropsy"

(i) Caloric deficiency; (ii) Protein deficiency and (iii) Water excess. Subsequently much work has been done on the relationship between the plasma proteins and the oedema accompanying malnutrition. MAVER(104) in a full discussion of war dropsy comes to the conclusion that it is a deficiency disease, the result of protracted existence on a diet deficient in total calories, especially protein. SCHICK and WAGNER (130) reported two cases in children of oedema arising from under-nourishment, though they do not ascribe the oedema to protein deficiency alone, while WIECHERS(154) reported a case of intestinal tuberculosis in a child of six which was accompanied by oedema which he ascribed to lack of absorption from the intestinal tract.

POYNTON and MACGREGOR (121) reported a case of gastro-jejuno-colic fistula accompanied by oedema and conclude by saying that "the one common condition in all these cases is semi-starvation, the result of either lack of food or non-absorption due to gastro-intestinal disturbances. This man was receiving an adequate mixed diet but owing to the short-circuiting of the small intestine, food could not be absorbed." In a case /

case of tuberculous enteritis, who developed oedema of the face, thighs, legs and sacrum and ascites after treatment with a markedly restricted diet, LANDIS and LEOPOLD (88) found the plasma proteins to be only 3.6 gm. per cent. with an osmotic pressure of 12.0 mm. of mercury.

BRUCKMAN, D'ESOPPO and PETERS (21) found that in one hundred and twenty-four determinations on fifty-seven patients who showed evidence of malnutrition the total serum proteins were reduced, the reduction being entirely in the albumin fraction. They conclude "the invariable finding of serum albumin deficiency in patients who have become malnourished as a result of a variety of diseases, and the demonstration that the serum albumin rises when protein is stored and nutrition improves, point strongly to the fact that protein starvation regularly causes the serum albumin to fall." In a further series of cases BRUCKMAN and PETERS (22) found that in patients with malnutrition, oedema almost invariably developed when the serum albumin fell below 3 gm. per cent., and was seldom found when the albumin exceeded 4 gm. per cent.

WEECH and LING (149) in a series of patients suffering from nutritional oedema, studied both during active and convalescent stages, found that when the level /

level of serum albumin was above 2.9 gm. per cent. oedema was never observed, and when the level fell below 2.5 gm. per cent. oedema was invariably present. As a result of metabolic observations on two of the patients they suggest that increase in the intake of sodium beyond the amount which can be excreted leads to retention, which in turn is accompanied by retention of Cl. and water to maintain the normal composition of the body fluids. Whatever the mechanism which produces this effect it would appear to bear a close relation to the serum protein level, and it is tentatively suggested that the threshold for the excretion of Na. may be controlled by the serum protein level.

The results reported by YOUNG, BELL, DONLEY and FRANK (157) are of especial interest in that many of their cases were mild ones and all were ambulant. In spite of this, they also found in a series of thirty-one patients that the serum albumin was slightly or moderately reduced, the globulin normal or increased and the calculated osmotic pressure slightly or moderately reduced. The difference between these results and those already reported is probably due to, among other factors, the comparative mildness of the condition and the fact that the patients were ambulant, because /



because, as DRURY and JONES (36) have pointed out, the erect human being is always near oedema owing to the comparatively small margin of safety between the capillary blood pressure and the onkotic pressure of the blood.

On the experimental side HARDEN and ZILVA (60) demonstrated the development of oedema in monkeys kept on a daily diet of 250-300 gm. of polished rice and yeast. KOHMAN (78) showed that rats given a diet adequate in every respect except that it was deficient in proteins invariably developed oedema which in turn disappeared when sufficient protein was added to the diet. In one rat oedema was produced and cured three times by alternating a low-protein diet with an adequate protein diet (76). Similar results were obtained by FRISCH, MENDEL and PETERS (55A).

The most recent contribution to the subject has been that of BLOOMFIELD (13) who reports that rats placed on a low protein diet for twenty-one weeks, in spite of marked loss of body weight, showed no significant decrease of serum protein concentration. In explanation of the discrepancy between his results and those of other workers he states that in the diets of other workers carrots were one of the principal features, while he found that a diet of carrots alone leads /

leads to a definite fall in serum protein concentration. "May these vegetables", he asks, "and perhaps other articles of food contain some agent which is antagonistic to the formation of blood proteins?" Interesting though these results may be, they will require confirmation, particularly in view of the fact that KOHMAN (78) had already shown that the oedema does not develop as a result of some toxic substance in the carrots, by demonstrating that rats fed on the control diet with the same carrot content as the low protein diet but with adequate protein did not develop oedema.

BLOOMFIELD's work indeed only emphasised the generally accepted thesis that there are several factors contributing to the onset of nutritional oedema, including a high water and salt intake, an acid diet, a possible increase in capillary permeability and the possibility of some interference with the regeneration of the serum proteins. SHANKS and DE (134) for instance in reporting the pathological findings in four cases of fatal epidemic dropsy, found that the chief feature was an extreme and widespread dilatation of the capillaries. What does seem to evolve very definitely from a study of the literature is that the serum protein level plays the dominating rôle in deciding the onset of oedema, and it is for this reason /

reason that the subject has been considered so fully here.

### Renal Oedema.

The findings of other workers have already been referred to (p.19), from which it is evident that the general consensus of opinion is that in renal oedema there is a distinct diminution in the plasma proteins, and it is only necessary now to discuss these findings in their relation to the formation of oedema.

LEITER (91A) in his exhaustive monograph on "Nephrosis" is firmly convinced that in this disease the sequence of events is:-

Albuminuria;  $\Rightarrow$  Low Plasma Proteins;  $\Rightarrow$  Reduced Osmotic Pressure of Plasma Proteins;  $\Rightarrow$  Nephrotic Oedema.

"The clinical evidence for this sequence is so overwhelming as scarcely to admit of any other interpretation than one of cause and effect." The evidence of course is that the albuminuria in these cases is so intense as to account for the hypoproteinaemia and that the oedema fluid has such a low protein content as to account for only a fraction of the protein that is lost from the blood. The same might be considered to hold true for the nephrotic type of glomerulonephritis where the findings are very similar to those in nephrosis, /

nephrosis, though the reduction in the plasma proteins may not be so marked nor the protein content of the oedema fluid quite so low, and for the present purpose the two will be considered together.

This conception of the aetiology of renal oedema is not universally accepted in its entirety, and some workers insist upon the necessity for some further factor in addition to the lowered onkotic pressure of the blood. The commonest conception of this additional factor is an increased affinity of the tissues for water, and it is stated by those who support this conception that there is not an exact parallellism between the plasma protein level and the degree of oedema. ALDRICH and M'CLURE (3), for instance, in discussing nephrotic oedema reject the view that the tissues are passive in oedema and believe that "the tissues take an active part in the formation of these oedemas", this being due to a general intoxication resulting from infection and causing a change in the tissues which increases their avidity for water.

TAREEV and his co-workers (144) conclude that in renal diseases the lowering of the onkotic pressure by the blood proteins is parallel to the rise of hygroscopic capacity of the tissues and the degree of /

of oedema, and rather tend to emphasise the part played by the tissues. KUMPF (77) in a study of the oedema produced in rabbits by prolonged bleeding states "It is difficult to explain the sudden onset of the oedema in rabbits on the basis of an altered concentration of plasma protein or of albumin. An increased affinity for water on the part of the tissue colloids would help to explain the transfer of water and the resulting oedema." It should be noted, however, that in his experiments there was a high protein content in the oedema fluid and as the hypoproteinaemia was not marked, it is doubtful whether the condition can be compared to nephrosis as it occurs clinically. Finally MEYER (110) is of the opinion that the colloid osmotic pressure of the blood is merely an expression of what is happening in the tissues and that repeated measurements of this pressure enables a conception to be formed as to the direction of the forces in the tissues which influence water regulation.

This conception of increased tissue avidity for water is one which has had many reincarnations, but the proof on its behalf is still lacking, while the evidence against it is quite definite. GOVAERTS (58) has shown that cellular tissue placed in plasma with a lowered /



lowered onkotic pressure does not show any more marked swelling than in normal plasma. SCHADE and CLAUSEN (129) found that nephrotic oedema fluid was actively absorbed through a dialytic membrane by the corresponding serum, in spite of the low onkotic pressure of the latter; they pointed out that oedema fluid, with a protein content lower than that of the blood, could exert no attraction for the fluid of the blood, and so would act as a buffer between the blood and tissues. In other words, even though such a thing as increased affinity of the tissues for fluids did exist, it would never draw water out of the blood in the presence of free oedema fluid.

A further development of the tissue affinity theory is that advanced by ALDRICH (2) who believes that, in nephrosis at least, a toxic substance elaborated as the result of infection so affects the tissue cells that their affinity for water is increased, and that oedema is a protective function in that it dilutes the toxins in each cell, in support of which he states that in his cases, whenever it has been possible suddenly to relieve this intoxication, the attack of oedema has been cut short.

The apparent lack of correlation between the plasma protein level and the cause of the oedema has led many workers to advance hypotheses which would explain /

explain the discrepancy. One such theory is that advanced by CALVIN and GOLDBERG (24) who postulate "some 'trigger' mechanism of unknown origin," probably similar to the action of certain diuretics which suddenly causes the retention of water, a diuresis, resulting in the typical "cylic" oedema of the nephrotic syndrome in children. They agree that a low serum albumin is the important predisposing factor, but they are of the opinion that it cannot entirely account for the sudden changes in oedema which they found.

M'CLURE, DE TAKATS and HINMAN (114) also disturbed by the apparent lack of precise correlation between the plasma proteins and oedema suggested variation in blood volume as one of the possible factors. This, however, is contrary to the findings of LINDER, LUNDGAARD and others (95) who found no appreciable increase in the serum volume nor were the fluctuations of the serum proteins or the variation of oedema accompanied by comparable changes of serum volume. Similarly ROWNTREE and BROWN (124) found a "normovolaemia" in these cases. Another argument against alteration in blood volume being anything like an important factor is the fact that the albumin and globulin fractions seldom show the same variation, in many /

many cases, indeed, the globulin increasing while the albumin decreases. Such a finding cannot be explained on the basis of alteration in the blood volume.

In an attempt to obtain an accurate evaluation of the role of the plasma protein in nephrotic oedema LOEB and his co-workers (96) have investigated the responses of a normal individual on a "salt-poor" regime and of a patient with nephrotic oedema to the ingestion of potassium chloride, ammonium chloride and sodium chloride. Their conclusions are that the responses of nephrotic and normal individuals are qualitatively alike, and that the quantitative differences observed are probably to a large extent dependent on differences in serum protein concentration. They are thus satisfied that WIDAL's conception of specific ion disturbances of renal or tissue behaviour is not the determining factor in nephrotic oedema; and, while admitting the importance of serum protein concentration, they do not believe that the mechanism of nephrotic oedema can be solely explained by alterations in serum protein concentration - "until the physio-chemical laws governing the excretion of various ions are understood, an evaluation of the abnormalities present will remain difficult." This undoubtedly expresses the present position /

position - the predominating importance of the plasma protein concentration, modified in varying degrees by the other factors which play a part in the regulating of the water balance of the body. As MOORE and VAN SLYKE have expressed it "So long as the protein content (of the blood) is below the critical level we can be practically certain that relaxation of salt restriction will be followed by recurrence or increase of oedema." (111)

A factor, which some have considered of importance in the aetiology of nephrotic oedema, is the presence of lipaemia in these cases. STEPP (141) and STRAUSS (143) both reported a distinct correlation between the blood cholesterol and the presence or absence of oedema, while DYKE (37) found that the deposition of cholesterol esters was the one histological factor common to the kidneys in six cases of nephritis of the "subacute parenchymatous" type, all associated with intense oedema. <sup>(105)</sup> MAXWELL reported that renal oedema is almost invariably associated with an increase in plasma cholesterol, and that the cholesterol figures fell as the oedema disappeared. He did not consider, however, that the two were related as cause and effect, but that they were both the result of some toxic process acting upon the tissue /

tissue cells, particularly those of the reticulo-endothelial system. HILLER and his co-workers, (66) however, considered the increase to be due to some disturbance of the balancing mechanism whereby lipids are much more slowly removed from the blood than under normal conditions.

This lipaemia is also found experimentally after haemorrhage in animals (FISHBERG (53), KUMPF (77) etc.) and it has been suggested that it may be correlated with an effort of the organism to compensate for the decrease in osmotic pressure following the loss of serum protein (FISHBERG). This, however, has not been confirmed and CALVIN and GOLDBERG (24) who, in children presenting the nephrotic syndrome, found in general that as the oedema increased the cholesterol increased and the albumin decreased, are not prepared to subscribe to the idea that the increased blood cholesterol is a compensatory mechanism in view of the variations observed. There is thus no direct evidence that the lipaemia has any relation to oedema, and on the whole HILLER's suggestion seems the most probable.

As to the cause of the protein deficiency in these cases it is now generally agreed that three factors are involved - (i) Loss in the urine;  
(ii) /



(ii) Interference with the mechanism of production;  
 (iii) Dietary insufficiency. As to the relative importance of each of these factors there is inevitably some difference of opinion. "Probably two factors responsible for changes in plasma protein concentration in nephritis - (i) The more important - a disturbance in the mechanism of their production, consequent upon the nature of the disease, the presence of infection or toxaemia, and dietary vagaries; (ii) Loss in the urine of large amounts of protein, especially albumin". (LINDER, LUNDSGAARD and VAN SLYKE (94)).

"The general tendency towards an inverse relation between plasma and urine ratios would seem to point to excretion of one type of protein in the urine as at least a partial explanation for the loss of protein in the plasma. In view of the deviations however, it appears that other factors in addition to protein excretion are involved in the process of lowering the plasma proteins". (HILLER, M'INTOSH and VAN SLYKE (67)).

"Malnutrition, or more specifically protein starvation, may be a factor in the production of serum albumin deficiency in oedematous nephritis. .

. . . Besides albuminuria, which permits the direct /

direct loss of serum albumin into the urine, the chief cause of serum protein deficiency appears to be depletion of the protein stores of the body". (PETERS et al.(119))

The question inevitably arises as to the possibility of replacing the missing plasma protein by an increase in the protein-intake. EPSTEIN (44) recommended massive amounts of protein - 120-240 gm. daily. PETERS and his co-workers (119) did not give such large amounts - seldom more than 125 gm. daily, finding that the amount was limited less by the physician than by the appetites and digestions of the patients. They report that when the patients were afflicted by any complicating wasting disease and were able to consume over long periods enough protein to supply the endogenous metabolic needs as well as sufficient to restore that lost in the urine and a further additional amount to restore the previously depleted tissues, the serum proteins rose and the oedema disappeared. At no time did they find any evidence of high protein diets increasing the proteinuria. In nephrosis LEITER (91A) recommends ample diet, but considers EPSTEIN's diet unpalatable.

Such a high protein diet is rather counter to the orthodox treatment of nephritis, but there is no evidence /

evidence that it in any way increases the renal damage. WORDLEY (156) found that an increased protein content of the diet did not increase the albuminuria, while M'CANN (113) states quite definitely that "liberal protein allowances in the diet do not of themselves injure the kidney," recommending that full advantage should be taken of the tendency to deposit protein by all individuals who have lost it either by inanition or by tissue destruction, and pointing out that protein that is deposited makes no demand on the excreting function, but builds up depleted tissue and circulating proteins with beneficial effects in the course of the disease. KEUTMANN and M'CANN (75) in four patients with chronic haemorrhagic Bright's disease, each receiving 40-200 gm. of protein daily, observed no deleterious effects upon the course of the disease; rather was there general clinical improvement in all cases with increasing weight (not due to oedema). They also state that the serum proteins fluctuated independently of the level of protein intake.

It is thus evident that in nephritis there is no contra-indication to a high protein diet, while in cases exhibiting the nephrotic syndrome there is good evidence for believing that such a diet is one of the main /

main factors in ensuring a disappearance of the oedema.

#### Cardiac Oedema.

The rôle of the plasma protein deficiency in the aetiology of cardiac oedema is one which has received comparatively little attention in the past, but the work that has been published is all very suggestive. EPSTEIN (42) reported twelve cases which he divided into two groups. In four of the cases the plasma proteins were only slightly diminished, the albumin fraction varying from 3.49 to 4.98 gm. per cent., and the globulin 1.99 to 2.54 gm. per cent. None of them apparently had any oedema and he adds "A moderate state of dilution or hydraemia would account fully for these values." In the other eight there was a greater diminution in the total proteins, and the globulin was less diminished than the albumin. Two of this latter group were cases of chronic myocarditis with anasarca, and the figures for them were respectively - Albumin 1.81 gm. per cent., Globulin 1.93 gm. per cent., and Albumin 1.37 gm. per cent. and Globulin 2.04 gm. per cent. Of the first group he says there is a moderate dilution of the blood (true hydraemia); the proteins retain their normal /

normal proportions. The second group he sums up by pointing out that the total proteins are moderately or markedly reduced, especially the albumin, so that the globulin may predominate and there is oedema. The change in the quantitative relations of the serum protein is not due to hydraemia. In a later publication (44) he gives the average composition of the blood in "cardiac condition" as - Albumin 4.42 gm. per cent. Globulin 2.24 gm. per cent., compared with his "normal" of Albumin 4.66 gm. per cent., Globulin 2.74 gm. per cent. It is thus evident that in this latter case he has not divided his cardiac cases into an oedematous and non-oedematous group.

One of the most comprehensive studies has been that of GOVAERTS. In his first paper (56) he says "l'oedème de stase", the result of local circulatory trouble or cardiac inefficiency, when moderate, may not be accompanied by any alteration in the osmotic pressure of the proteins. But this is not the rule, and oftener, when such oedema is present, the osmotic pressure of the proteins is low. Every time the osmotic pressure due to the proteins was below 30 cm. (of water) oedema was present." In his "cardiopathie avec néphrite de stase", with marked oedema the onkotic pressure was low - 25.5-16.5 cm. of water as compared with a "normal" of 35-40 cm. of water. In a /



a subsequent paper (58) he confirms these results and gives one case of mitral disease and auricular fibrillation with marked oedema in which the onkotic pressure was 21.6 cm. of water, the albumin being 1.73 gm. per cent., and the globulin 4.83 gm. per cent.

COPE (31), who included a few cardiac cases in his study of nephritic oedema, reported that the plasma protein osmotic pressure in oedema from myocardial failure was only slightly reduced, and was markedly higher than in cases presenting a comparable degree of oedema of renal origin. His series includes eight cases of cardiac oedema and the onkotic pressure varied from 20.2 to 39.9 cm. of water, compared with his normal of 30-32 cm. of water.

IVERSEN and NAKAZAWA (71) in a larger series, were of the opinion that in patients with cardiac oedema there is always a lowering of the onkotic pressure. Their figures varied from 17.3 to 36.3 cm. of water, there being only two cases over 30.0 cm. of water, as compared with a normal figure of 32.5-40.1 cm. of water.

PAYNE and PETERS (117) in a separate study of the serum proteins in heart disease, which included sixteen cases with oedema found that the albumin varied from 2.89 to 5.83 gm. per cent. and the globulin from /

from 1.17 to 2.94 gm. per cent., and in every case except one the total protein concentration was less than 7.0 gm. per cent.

MEYER (110) and EPPINGER (41) also reported a lowering of the colloid osmotic pressure in cardiac oedema, and EPPINGER emphasised the increase in the globulin fractions.

On the other hand there is the very definite but entirely unsupported statement of MOORE and VAN SLYKE (111) in the course of a discussion on nephrotic oedema that "It is well known that the oedema of heart failure is not attributable to plasma protein deficiency", while TAREEV and his co-workers (144) also state that in cardiac decompensation oedema is not accompanied by a drop in the onkotic pressure of the plasma proteins.

It is thus evident that what work has been done on the subject is in agreement with our own results, which show that in cardiac oedema there is a distinct fall in the plasma protein concentration. That this diminution is not merely an accompaniment of heart disease is shown by a comparison of the results in the group of cardiac cases with oedema and those without oedema, for, whereas in the cases with oedema 91 per cent. have an onkotic pressure less than 29 cm. of water /

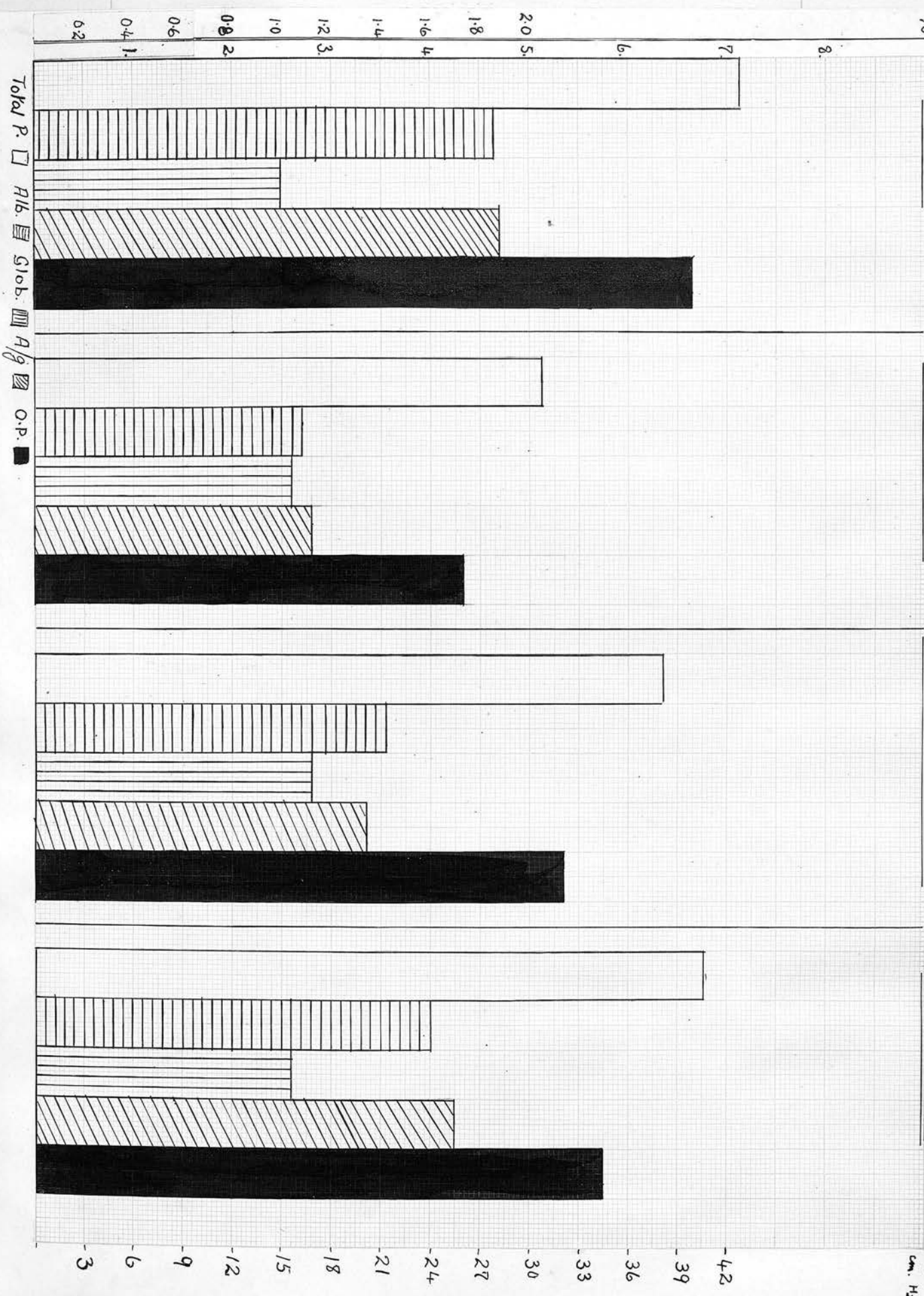
A/g Rat. Normals.

Cardiac Oedema.

Cardiac CT Oedema.

Miscellaneous.

Fig. X.

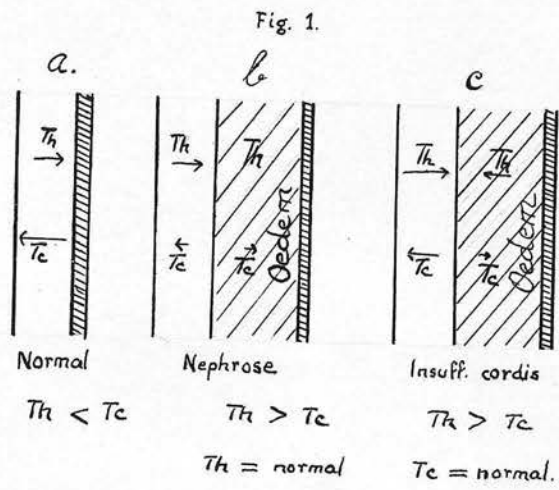


water, of the cases without oedema 91 per cent. have an onkotic pressure greater than 29 cm. of water. The position is shown graphically in Fig. X which records the average results in the two cardiac groups, the normal group and the miscellaneous group, and from which it can be seen that of these four groups the most striking fall in the plasma protein concentration is in the "cardiac oedema" group, with a smaller diminution in the "cardiac without oedema" group, while the "miscellaneous" group occupies a position between the non-oedematous cardiac group and the normal figures. If Fig. X in turn be compared with Fig. VII it is seen that the figures for the "cardiac oedema" group are higher than for the cases of subacute nephritis with oedema.

Having thus established that there is a definite fall in the plasma protein concentration in cases of heart disease with oedema, the two problems which arise are the significance of this fall in the aetiology of the oedema and its cause.

The /

Fig. XI.





The Significance of the Low Plasma Protein  
Concentration in Cardiac Oedema.

The more important factors in the causation of cardiac oedema have already been discussed as well as the emphasis laid upon them by different workers, and from such a discussion there evolves the well-known fact that no one factor plays a dominant rôle in the causation of such oedema, the importance of any one factor varying from case to case. The importance of the plasma proteins lies in their relation to the capillary pressure.

As has been shown one of the main factors in the control of fluid exchanges between the bloodstream and the tissues is this balance which exists between the hydrostatic pressure tending to drive fluid into the tissues and the onkotic pressure tending to draw fluid into the bloodvessel. This balance can naturally be upset in one of two ways, either of which will result in an increased passage of fluid into the tissues - (i) The hydrostatic pressure may be raised, the onkotic pressure remaining unaffected; (ii) The onkotic pressure may be lowered, the capillary pressure remaining normal. Between these two extremes any modification may occur. Fig. XI which is reproduced from a paper by IVERSEN and NAKAZAWA (71) represents /

represents this diagrammatically. In nephrotic oedema there is a marked fall in the onkotic pressure, the capillary pressure remaining comparatively unaffected, while in cardiac oedema there is a fall in the onkotic pressure, though not so marked as in nephrotic oedema, as well as a rise in the capillary pressure. In both, though attained by different methods, there is a preponderance of hydrostatic pressure over onkotic pressure, the result of which can only mean oedema.

This explains why the plasma protein, and consequently the onkotic pressure, are at a higher level in cardiac than in nephrotic oedema. It is also one of the reasons why anomalous cases of cardiac oedema may occur where there is only a very slight diminution in the plasma proteins, as in these cases there is such a marked rise in the capillary pressure that it only requires a slight fall in the onkotic pressure to result in such a preponderance of the hydrostatic pressure that oedema occurs.

That such a view is widespread is manifest from the following quotation:-

- (i) "Dans toutes les variétés d'oedèmes chroniques que nous avons étudiés, dans l'oedème de stase, dans les oedèmes cardiaques, dans les oedèmes renaux, les facteurs immédiats qui provoquent l'infiltration /

l'infiltration interstitielle sont toujours les mêmes. L'effet mécanique d'une pression intracapillaire supérieure à la pression osmotique des protéines fournit dans tous les cas une explication très satisfaisante de l'oedème". (GOVAERTS (56)).

(ii) The reduction of the onkotic pressure can be so great as in itself to explain the onset of oedema, but in all cases the increased hydrostatic pressure plays a definite, and in some cases the essential rôle. (IVERSEN and NAKAZAWA (71). IVERSEN (70))

(iii) In congestive heart failure, the increased hydrostatic pressure within the capillaries plays a comparatively small part, as there is usually a simultaneous reduction of the colloid osmotic pressure. (EPPINGER (41))

While these views may lay an undue stress on the importance of the capillary pressure in cardiac oedema, they do reveal the emphasis which has been laid on the reduction of the colloid osmotic pressure in the causation of the oedema. The other factor which undoubtedly plays a part is that of capillary permeability which, it is practically certain, is increased /

increased in these cases, but does not minimise the importance of the onkotic pressure.

The Cause of the Plasma Protein Deficiency  
in Cardiac Oedema.

Blood Volume.

It has been suggested that the fall in the plasma proteins is more apparent than real, being largely accounted for by an increase in blood volume. Such a view, however, is incompatible with the findings that, not only is there no correlation between the fall in the two protein fractions, but in the majority of cases there is an actual increase in the globulin fraction, the fall being entirely in the albumin fraction. Such a variation in the protein fractions could not be caused by a simple increase in blood volume. Further it is by no means decided yet whether there is a condition of hydraemia in cardiac oedema in spite of BOLTON's (18) findings of hydraemic plethora developing into true plethora and ROWNTREE and BROWN's (124) reports of a "simple hypervolaemia". STRAUSS (142), for instance, was of the opinion that hydraemia occurs only in severe cases /

cases of cardiac decompensation; VEIL and his co-workers (147) only found hydraemic plethora occasionally in their cases; BECKMANN (10) concluded that the condition in the blood depended upon the degree of saturation of the tissues with water, while BOCK (14) could find no change in the amount of blood plasma in cardiac disease.

#### Albuminuria.

In view of the important part played by albumin in the depletion of the plasma proteins in renal oedema, it might be considered that a similar factor was responsible for the depletion in cardiac oedema. For this, however, there is no evidence. In none of our cases was the albuminuria extensive enough to account for the hypoproteinaemia, and in many of the cases there was never more than a trace of albumin in the urine.

#### Transudation.

It is generally recognised that the protein content of the oedema fluid is higher in cardiac oedema than in nephrotic oedema, and might therefore be considered a possible explanation of the loss of protein from the blood. In nephrotic oedema such a possibility is definitely excluded as the protein content of the fluid is so extraordinarily small.

The /



The figures in our cases, however, (p. 24) are not sufficient to account for more than a fraction of the protein depletion of the blood.

#### Malnutrition.

In every case of cardiac decompensation there is a degree of malnutrition, the exact degree varying with the duration and severity of the illness. It has already been shown that in malnutrition and cachexia, no matter what the cause, there is a tendency towards a fall in the level of the plasma proteins, and in nephritic oedema it is generally accepted that this is one of the factors in causing the plasma protein depletion. This malnutrition in cardiac disease is due partly to the disease itself, partly to the lack of sufficient protein intake and partly to interference with protein formation. In view of the connection which has been shown to exist between the liver and protein formation it is not impossible that the chronic congestion of the liver, which is such a common feature of cardiac decompensation, may actually interfere with protein regeneration. It is thus probable that this is the dominant factor in causing the plasma protein depletion in cases with cardiac oedema. A similar conclusion is reached by PAYNE and PETERS (117) who tried to obtain an /

an objective measurement of the degree of malnutrition by obtaining the weight of the patient before the illness, during the illness, and after the oedema had disappeared - "The serum albumin deficits are due to malnutrition. The histories leave no doubt that anorexia is the chief cause of the malnutrition with nausea and vomiting frequently acting as contributing factors.

THERAPEUTICS. /

THERAPEUTICS.

There is no disease more chronic in its course and resistant to treatment than certain cases of cardiac oedema. The entire resources of the physician may be exhausted with no result apart from an occasional temporary subsidence of the oedema following upon the use of some particularly potent drug, a subsidence which is all too often followed by a recrudescence of the dropsy to a degree far in advance of that present prior to treatment. As the oedema, itself a symptom of congestive heart failure, throws an extra load on the heart and is thus a secondary further cause of "heart failure" (HARRISON and PILCHER (60A)) it follows that this failure to reduce the amount of oedema fluid must be an important factor in increasing the gravity of the prognosis, increasing as it does the load on the already overburdened heart. In addition there is the mental factor to be considered, as the water-logged patient, watching the enthusiastic yet unavailing efforts of his physician, finds his condition becoming steadily worse and finally becomes convinced that recovery is not to be.

The findings recorded here suggest a possible aid to /

to the alleviation of this condition. It has been part of the traditional treatment of cardiac decompensation to give the patient a very light diet, and particularly one which has a low protein content. The successful results obtained in the treatment of nephrotic oedema by means of a high protein diet, however, suggest that, if a plasma protein deficiency is one of the factors responsible for cardiac oedema, a high protein diet may also be of use in the alleviation of cardiac oedema. It may well be that many of these chronic cases of cardiac oedema in the past, which have proved so resistant to treatment, have really been accentuated by the treatment they have received, in that the malnutrition from which they have been suffering has been steadily progressing and so increasing the plasma protein deficiency which has thus come to occupy a more important part in the causation of the oedema. If such a patient had been given a diet containing the maximum of protein, in addition to the other treatment, there might very well have been a speedier and more satisfactory response to the treatment. "The common practice of restricting diet, and especially protein, in heart failure, may represent misdirected effort". (PAYNE and PETERS (117))

Bearing in mind the digestive disturbances almost invariably /

invariably present in these cases, patients with cardiac oedema should be given a diet containing the maximum of protein compatible with the digestive powers of the patient. Such a diet, containing an adequate portion of protein and yet assimilable by the patient with cardiac oedema, can be easily obtained, and would without doubt prove of great value in helping to reduce the oedema in many cases. Even though there may be a marked albuminuria it is not necessary to restrict the protein for, as has been shown in the discussion of renal oedema, there is no evidence that a high protein diet has an injurious effect on the kidneys. It is scarcely necessary to add that in such a diet the same restriction must be observed as to salt and fluid intake as are at present observed.

#### CONCLUSION /



### CONCLUSION.

One of the factors in the causation of cardiac oedema is a depletion of the plasma proteins which results in a fall in the colloid osmotic pressure of the blood. This depletion is due principally to malnutrition, and it is suggested that cases of cardiac oedema should be given diets containing the maximum amount of protein compatible with the digestive functions of the patient.

### SUMMARY /

SUMMARY.

1. The results are recorded of an investigation of the plasma proteins in sixty-five patients, consisting of twenty-three cases of heart failure with oedema, sixteen cases of heart disease unaccompanied by oedema, eight cases of Bright's disease, five cases of tuberculosis, two cases of ovarian carcinoma with gross ascites, a miscellaneous group of eight cases and three junior members of the hospital staff who were used as "normals". In all ninety-one estimations were made.
2. The protein content of the oedema fluid in these cases was estimated on twenty-nine samples.
3. There is a distinct diminution in the plasma proteins in cardiac oedema, 87.5 per cent. of the cases having a plasma albumin content of less than 3.2 gm. per cent., while 91 per cent. have a colloid osmotic pressure of the blood below 29 cm. of water.
4. In heart disease without oedema there is only a slight diminution in the plasma proteins as compared with the normal, 87 per cent. of the cases /

cases having a plasma albumin level greater than 3.2 gm. per cent., and 91 per cent. having a colloid osmotic pressure greater than 29 cm. of water.

5. In view of these findings it is suggested that plasma protein deficiency plays an important role in the aetiology of cardiac oedema.
6. The origin, functions and variations of the plasma proteins are discussed in relation to their role in the production of oedema.
7. The main cause for the plasma protein depletion in cardiac oedema is shown to be malnutrition.
8. In view of this latter finding it is suggested that the dietary of patients with cardiac oedema should contain the maximum amount of protein compatible with their digestive powers.

A C K N O W L E D G M E N T S.

I have to express my indebtedness to Professor W. T. Ritchie for permission to investigate the cases in the wards under his charge in the Royal Infirmary, Edinburgh, and to Professor D. Murray Lyon for the full facilities offered to carry out the biochemical investigation in the Clinical Laboratory of the Royal Infirmary, Edinburgh.

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A P P E N D I X.

0.98.    Male.    Aet. 58.    Myocardial Failure.    Fig.II.

13.8.31    Admitted to surgical ward and operated upon at once on account of a perforated duodenal ulcer.

No previous history of rheumatism, venereal disease or cardiac disability.

28.8.31    Discharged to Convalescent Home, having made satisfactory recovery.

30.8.31    Confined to bed on account of dyspnoea and swelling of legs.

1.9.31    Admitted to medical ward.

Patient orthopnoeic.    Oedema of lower limbs  
Fluid in right pleural cavity.    Crepitations  
audible at bases of both lungs.

Heart:- slightly enlarged.    Heart sounds  
feeble but pure.    Temperature:- 99.2° F.

Pulse Rate:- 112 per minute.

4.9.31    Right pleural cavity aspirated - 1020 c.c.  
withdrawn.

16.9.31    35 c.c. fluid obtained from right leg.  
Oedema diminishing slowly.

Electrocardiogram:- low voltage record.

18.9.31 /



18. 9.31      Neptal, 1 c.c. given intramuscularly.  
19. 9.31      "      1 c.c.      "      "  
20. 9.31      "      1 c.c.      "      "  
21. 9.31      "      1 c.c.      "      "  
3.10.31      No pitting of lower limbs.      Still some  
                  fluid in right pleural cavity.  
17.10.31      Patient allowed up for a quarter of an hour.  
9.11.31      Patient discharged from hospital, being  
                  able to indulge in light exercise.

Urine:-      There was never at any time more  
                  than a trace of albuminuria.

0.149.    Male.    Aet. 55.    Myocardial Failure.    Fig.III.

13.10.31    Admitted to hospital with history of  
                  increasing dyspnoea on exertion for a year.  
                  Swelling of the legs had been noticed during  
                  the preceding fortnight, during which period  
                  patient had also had a cough.      Confined to  
                  bed for six days.

                 History of rheumatic fever in 1898.  
                  Patient was orthopnoeic.      Gross oedema of  
                  both lower limbs, ascites and a slight  
                  effusion into the right pleural cavity.

Heart /

Heart:- Definitely enlarged. Gallop rhythm. Heart sounds pure but faint.

Temperature:- 97°F. Pulse Rate:- 120 per minute. Respiratory Rate:- 28 per minute.

Wassermann reaction, negative.

- 15.10.31 Electrocardiogram:- Low voltage with inversion of T wave in Leads II and III.
- 19.10.31 No improvement in oedema.
- 20.10.31 Neptal, 1 c.c. given intramuscularly.
- 22.10.31 " 1 c.c. " "
- 3.11.31 Oedema much improved and patient's general condition satisfactory.
- 12.11.31 Oedema now absent. Patient allowed up for a quarter of an hour.
- 10.12.31 Patient up for two hours.
- 16.12.31 Discharged from hospital very much improved and able to walk about quietly without any discomfort.

Urine:- Never more than trace of albumin.

0.230. Female. Aet. 52.

Fig. IV.

Myocardial Failure. Auricular Fibrillation.

- 15.12.31 Admitted to hospital with history of increasing dyspnoea on exertion and intermittent swelling of legs for a year. No history of rheumatic fever or miscarriages.
- Patient was orthopnoeic with gross oedema of both lower limbs. Ascites present and effusion in right pleural cavity.
- Heart:- Enlarged. Heart sounds faint but pure. Pulse Rate:- 100 per minute. Wholly irregular. Temperature:- 97.4°F.
- Respiratory Rate:- 28 per minute.
- Hepatic enlargement.
- Urine:- 8 gm. albumin per litre.
- 18.12.31 650 c.c. fluid aspirated from right pleural cavity.
- 21.12.31 Patient more comfortable but no diminution of oedema.
- 31.12.31 Increased hepatic enlargement.
5. 1.32 Oedema diminishing. Patient still breathless.
21. 1.32 Oedema still diminishing.
30. 1.32 Oedema increasing again. Cardiac delirium at nights.
1. 2.32 /

1. 2.32 Neptal, 1 c.c. given intramuscularly.
5. 2.32 Oedema still increasing. Patient weaker.
- 10.2.32 Neptal, 1 c.c. given intramuscularly.
- 11.2.32 " 1 c.c. " "
- 12.2.32 Incontinence of urine.
- 15.2.32 Neptal, 1 c.c. given intramuscularly.  
Oedema stationary. Patient weaker.
- 18.2.32 Oedema of right arm.
- 27.2.32 Oedema increasing steadily and patient's  
general condition very unsatisfactory.  
Allowed to return home at own request.

Urine:- Throughout her stay in hospital  
there was a persistent albuminuria which was  
never less than 2 gm. per litre.

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